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Sublethal Metabolic Responses of the Hermatypic Coral Madracis decactus Exposed to Drilling Mud Enriched with Ferrochrome Lignosulfonate

Texas A and M Univ., College Station

Prepared for

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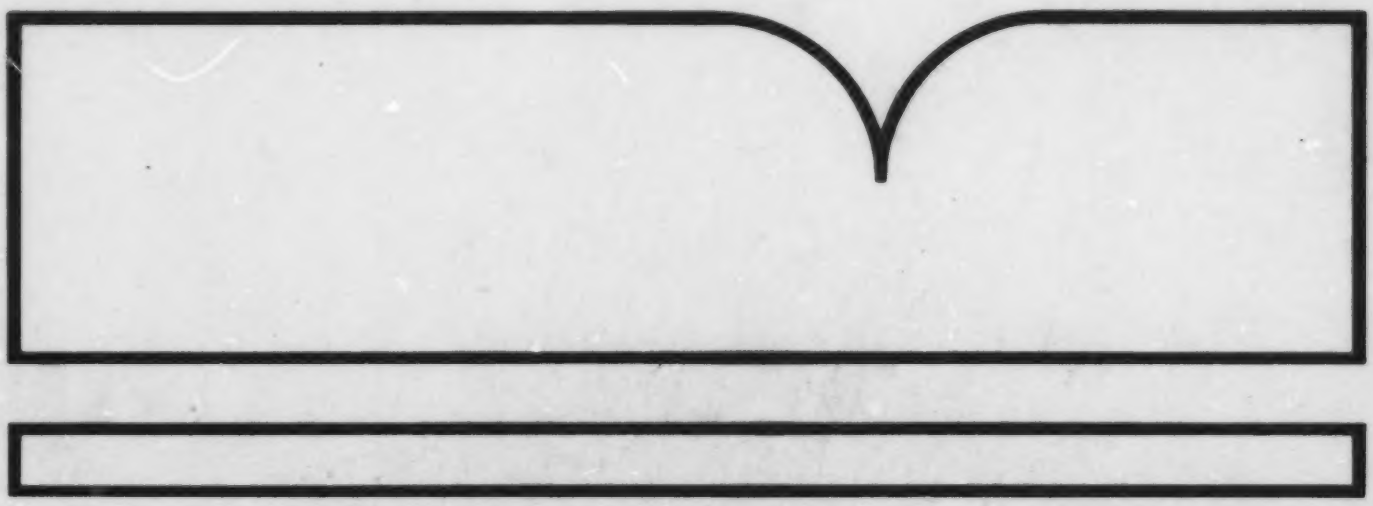

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Sublethal Metabolic Responses of the Hermatypic Coral *Madracis decactus* Exposed to Drilling Mud Enriched With Ferrochrome Lignosulfonate

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All corals exposed to FCLS reacted by reducing their polyp expansion behavior, although only the two showed mass polyp mortality. When exposure to drill mud + FCLS was discontinued, respiration and excretion of surviving corals remained low and stable while their polyp activity returned to normal levels within 48 hours.

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FOREWORD

The protection of our estuarine and coastal areas from damage caused by toxic organic pollutants requires that regulations restricting the introduction of these compounds into the environment be formulated on a sound scientific basis. Accurate information describing dose-response relationships for organisms and ecosystems under varying conditions is required. The Environmental Research Laboratory, Gulf Breeze, contributes to this information through research programs aimed at determining:

- the effects of toxic organic pollutants on individual species and communities of organisms;
- the effects of toxic organics on ecosystems processes and components;
- the significance of chemical carcinogens in the estuarine and marine environments.

Over the past decade, oil and gas drilling operations in the Gulf of Mexico have resulted in millions of cubic feet of drilling muds and other byproducts being released into the marine environment. This report describes a project to determine the sublethal effects of ferrochrome lignosulfonate, one of the principal drilling mud additives, to corals. The results of this study will be used to aid in the assessment of the potential impact of drilling mud discharge on marine organisms and ecosystems.

Henry F. Enos
Director
Environmental Research Laboratory
Gulf Breeze, Florida

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ABSTRACT

Madracis decactis corals were exposed for 17 days in laboratory aquaria to suspensions of 100 ppm drilling mud spiked with 0, 3, and 10 ppm ferrochrome lignosulfonate (FCLS). During the first week of exposure, these corals increased their oxygen consumption and ammonium excretion, relative to uncontaminated controls. Those corals exposed to the highest enrichments of FCLS demonstrated the greatest increases in respiration and excretion and also the largest variations in respiration and excretion between individual experimental animals. Corals reached their highest average rates of respiration and excretion by the end of the first week of continuous exposure. Rates then decreased during the next week and, after a secondary increase in excretion and respiration between days 10-13 which was most pronounced in those corals exposed to FCLS enrichment, leveled off at near-initial rates by the end of the second week.

Two corals, one exposed to 100 ppm drill mud + 3 ppm FCLS and the other to 100 ppm drill mud + 10 ppm FCLS, became noticeably moribund as the experiment entered its second week. These were the two corals which showed the most rapid and most consistent increases in excretion and respiration during the first week of exposure to FCLS. By week two, polyp expansion in both of these corals was dramatically reduced, and each was removed from the experiment when areas of bare corallum suggested the onset of polyp death.

All corals exposed to FCLS reacted by reducing their polyp expansion behavior, although only the two cited above showed mass polyp mortality. When exposure to drill mud + FCLS was discontinued, respiration and excretion of surviving corals remained low and stable while their polyp activity returned to normal levels within 48 hours.

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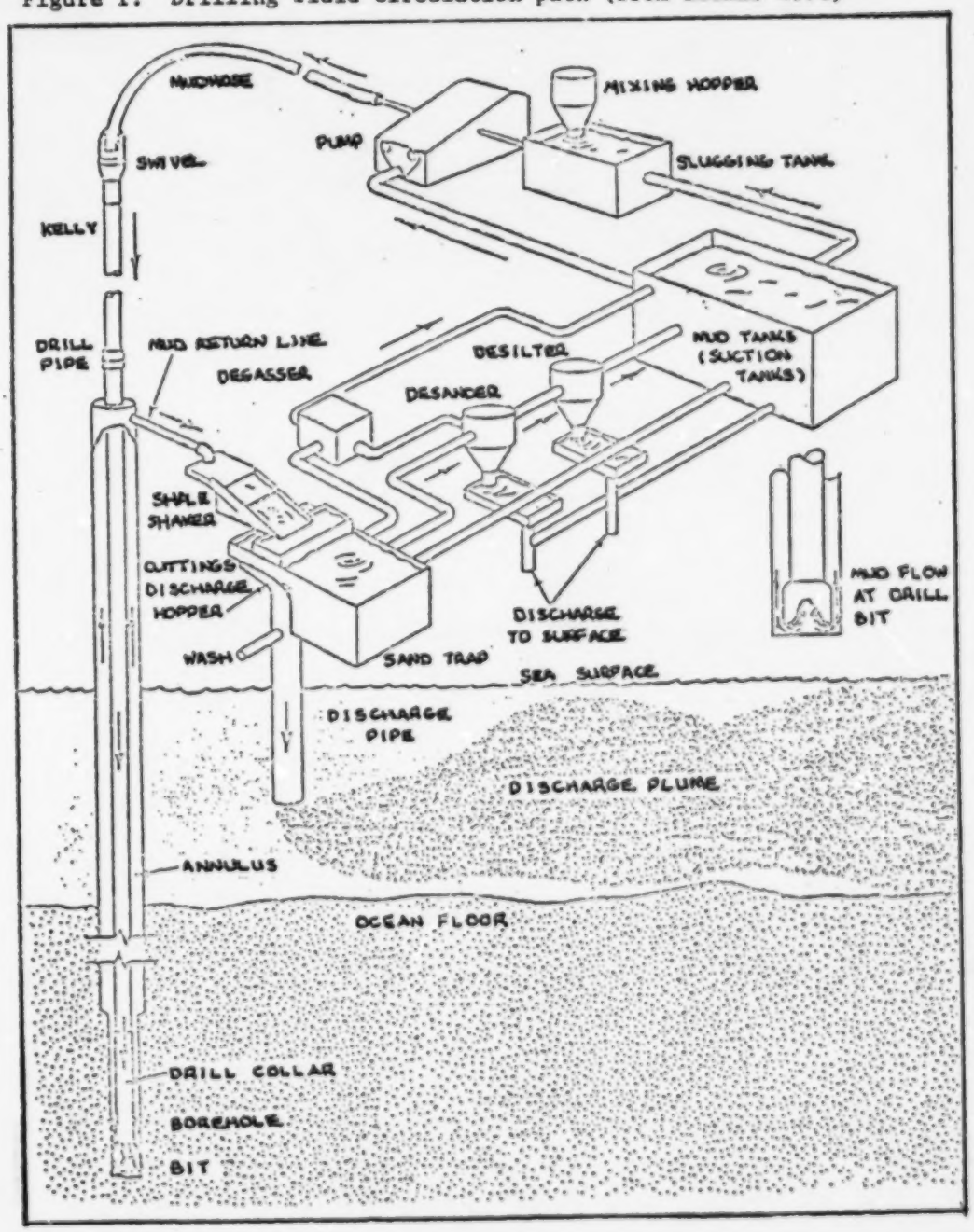
INTRODUCTION

Drilling Muds and Drilling Fluids. According to the U.S. Bureau of Land Management (1976), over 2700 oil and gas wells have been drilled since 1968 on the continental shelf of the Northwestern Gulf of Mexico. In ten years this level of drilling has produced literally millions of cubic feet of discharge drill mud and other drilling byproducts released into the environment. The Offshore Operators Committee (1978) estimated that 17,000 to 39,000 cubic feet of drilling mud and 11,000 to 22,000 cubic feet of cuttings are discharged during the drilling of a single offshore well.

Drilling mud is a slurry of barite and bentonite clays, water and chemical additives (American Petroleum Institute, 1978). Since muds are commonly prepared on the site at the drilling platform, the composition and proportions of each may be highly variable. According to Knox (1978), mud is pumped down the center of the drill bit, providing a stream of thick fluid to suspend the drilled chips at the bottom of the hole and to carry them to the surface (see Figure 1). The pumping pressure brings the mud and cuttings back up to the platform, where the cuttings are separated and mud returned to reservoirs for recirculation. Besides the primary function of suspending and removing cuttings, the circulation of drilling mud also cools and lubricates the bit, inhibits the loss of oil and gas, and helps to prevent the intrusion of water into the bore hole (Adler and Siegele, 1966).

The journal used as a pattern for format and style was Limnology and Oceanography.

Figure 1. Drilling fluid circulation path (from Ecomar 1978)



The very properties which make drill mud effective--density, gel strength, and viscosity--prevent dispersal by ocean currents (Knox, 1978). Relatively few studies have attempted to measure dilution rates and dispersion patterns of drilling fluids discharged from offshore drilling platforms. The present consensus is that rapid dilution occurs near the discharge point, with the rate of dilution decreasing as the discharge plume is advected farther from its source.

Flocculated clay spheroids appear to settle out most rapidly, while the finer cuttings form a suspended plume that is diluted by convection and turbulent diffusion as it moves away from the discharge pipe (Ray and Shinn, 1975; Zingula, 1975; Ecomar, 1978). The data presented in reports from Tanner Bank (Ecomar, 1978) and the Lower Cook Inlet (Miller, K.D., 1976) indicate that within 200 meters of the discharge point particulate concentrations of 100 ppm or greater may be expected. Their data indicate that beyond 200-500 meters from the discharge point, measured concentrations of suspended solids and the various drill mud additives (Ba, Cr, Pb) were indistinguishable from background levels.

Laboratory studies have demonstrated that the major components of drilling fluids (barite and bentonite) are relatively non-toxic to marine invertebrates (results summarized in McAufliffe and Palmer, 1976; Monaghan et al., 1976). However, ferrochrome lignosulfonate (FCLS), which is one of the principal additives used to thin the clay suspension, may be highly toxic to selected marine invertebrates. FCLS increases the pumpability by decreasing the gel strength and yield point, while it increases the chip carrying capacity by increasing the plastic viscosity (Kelly, 1964). The enrichment of drill mud with FCLS generally increases with depth of drilling.

Two theories have been proposed (see Knox, 1978) to explain the thinning ability of FCLS in clay suspensions. The first explains the clumping of clay particles by edge-to-edge attraction; at neutral and alkaline pH, the alumina-silica matrix has a net negative charge on its edges. The lignosulfonate complex, a polyanion, disrupts the attraction by absorbing to the edges of the particles and neutralizing the positive charge. The second theory is that the important attraction holding clay particles together is hydrogen bonding of water between the faces, and that lignosulfonate displaces this water and prevents the attraction. Neither theory accounts for the importance of chromium and iron to the thinning properties of FCLS.

In drilling mud the FCLS is not free, but adsorbed onto the clay particles (Knox, 1978). Consequently, instead of rapid dilution from effluents pumped into the water column, FCLS precipitates with the clay particles (Ecomar, 1978). Adsorbance onto the clay is reversible (Skelly and Dieball, 1970) so that FCLS is slowly released into the immediate vicinity of benthic inhabitants.

It is the chromium content of FCLS that is largely responsible for its toxicity. Chromium may occur in several oxidation states, the most common being hexavalent and trivalent. The hexavalent form is more soluble in seawater and more toxic to a wide variety of marine organisms than trivalent chromium (Mearns and Young, 1977). Marine organisms may accumulate chromium and other heavy metals by several processes. These include direct accumulation, either actively of passively by absorption and adsorption or indirect accumulation by ingestion of suspended material, sediments, and contaminated prey (Alexander et al. 1977).

Laboratory toxicity studies involving drilling muds have

concentrated primarily on acute responses of adult test organisms to individual drill mud components (see Table A-1 in Appendix). It is now generally accepted, however, that testing of isolated components of any complex mixture may be unrealistic. Moreover, few efforts have been made to relate test conditions such as concentration and exposure time to field conditions in the discharge area. In addition, laboratory estimates of long-term effects of sub-lethal concentrations of drilling muds on physiological processes and behavioral mechanisms are lacking.

FCLS Toxicity to Corals. Westerhaus (1978) demonstrated that dissolved FCLS was toxic to the coral Madracis mirabilis at concentrations of 50 ppm and 75 ppm within 96 hours of continuous exposure in flow-through laboratory aquaria. While experimental and individual variability were both quite high at these FCLS concentrations, her experiments suggested that coral respiration and ammonium excretion increased as a function of FCLS stress. Westerhaus postulated that these parameters of whole-animal metabolism might provide a readily measurable index of the relative "health" of corals stressed with drilling fluids.

The purpose of the present study was to expand upon Westerhaus' work by monitoring the oxygen consumption and ammonium excretion of corals exposed in the laboratory to lower, and presumably sublethal, concentrations of FCLS. Rather than testing FCLS independently, a 100 ppm suspension of drill mud was spiked with serial dilutions of 10 ppm, 3 ppm, and 0 ppm FCLS. These drill mud plus FCLS levels were selected to simulate loads which corals within 200 meters of the discharge might experience during an actual drilling operation. Corals were subjected to 17 days of continuous exposure to drill mud plus FCLS regimes to

contrast differences between short-term (1-7 days) and longer-term (greater than 2 weeks) responses.

MATERIALS AND METHODS

Coral Collection and Placement. All experiments were conducted at Stage-1, a three tiered 100' by 100' research platform located 12 miles offshore from Panama City, Florida. This offshore laboratory is managed by the Naval Coastal Systems Center. The laboratory seawater system intake at a depth of 29 meters provides oceanic water of $35^{o}/oo$.

The scleractinian coral species Madracis decactis was selected as a test organism since it is one of the most abundant coral species on Gulf of Mexico hard-bank reefs (Bright and Pequegnat, 1974). Colonies were collected at 29 meters using SCUBA from the Flower Gardens area of the Gulf of Mexico, which is the northenmost thriving tropical coral reef system in this region. This topographic high is situated approximately 107 nautical miles south of Galveston, Texas on the outer edge of the continental shelf at 93^{o} 48.5' west longitude and 27^{o} 52.3' north latitude (see Figure 2).

At the collection site the corals were placed in aerated IGLOO aquaria for transport to Freeport, Texas. Upon arrival the corals were transported by car to Panama City, Florida within 24 hours.

On stage the corals were placed on sea tables in the wet lab, where they were allowed to acclimate for 10 days to salinity, temperature, and natural particulate regimes of the Stage-1 seawater system. Coral heads were cleaved to appropriate size and then "groomed" by hand to remove epiphytic algae, small crabs, and other associated fauna so that these commensuals would not contribute to subsequent metabolic measurements. Corals were then placed in flow-through one-liter experimental aquaria

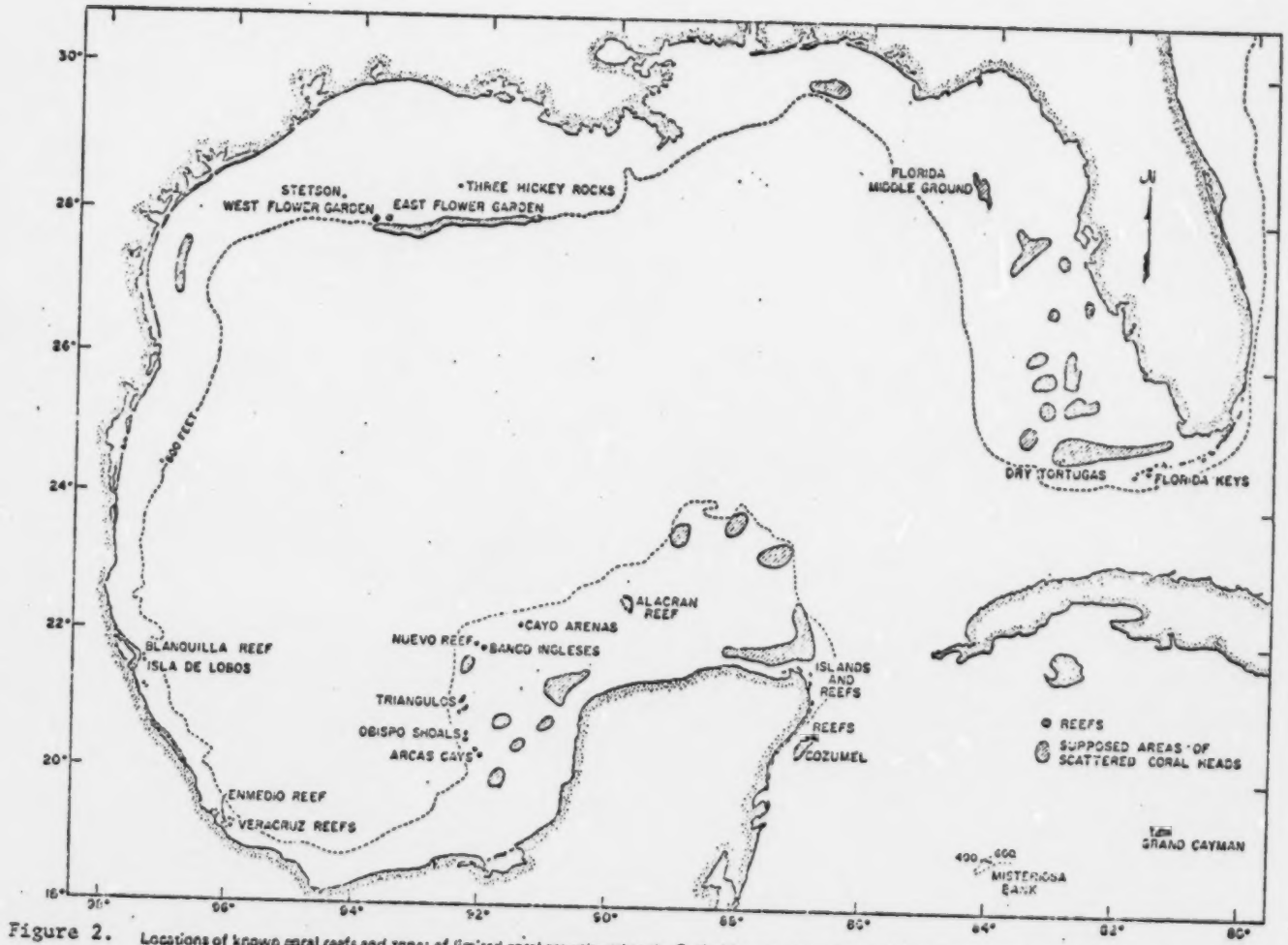


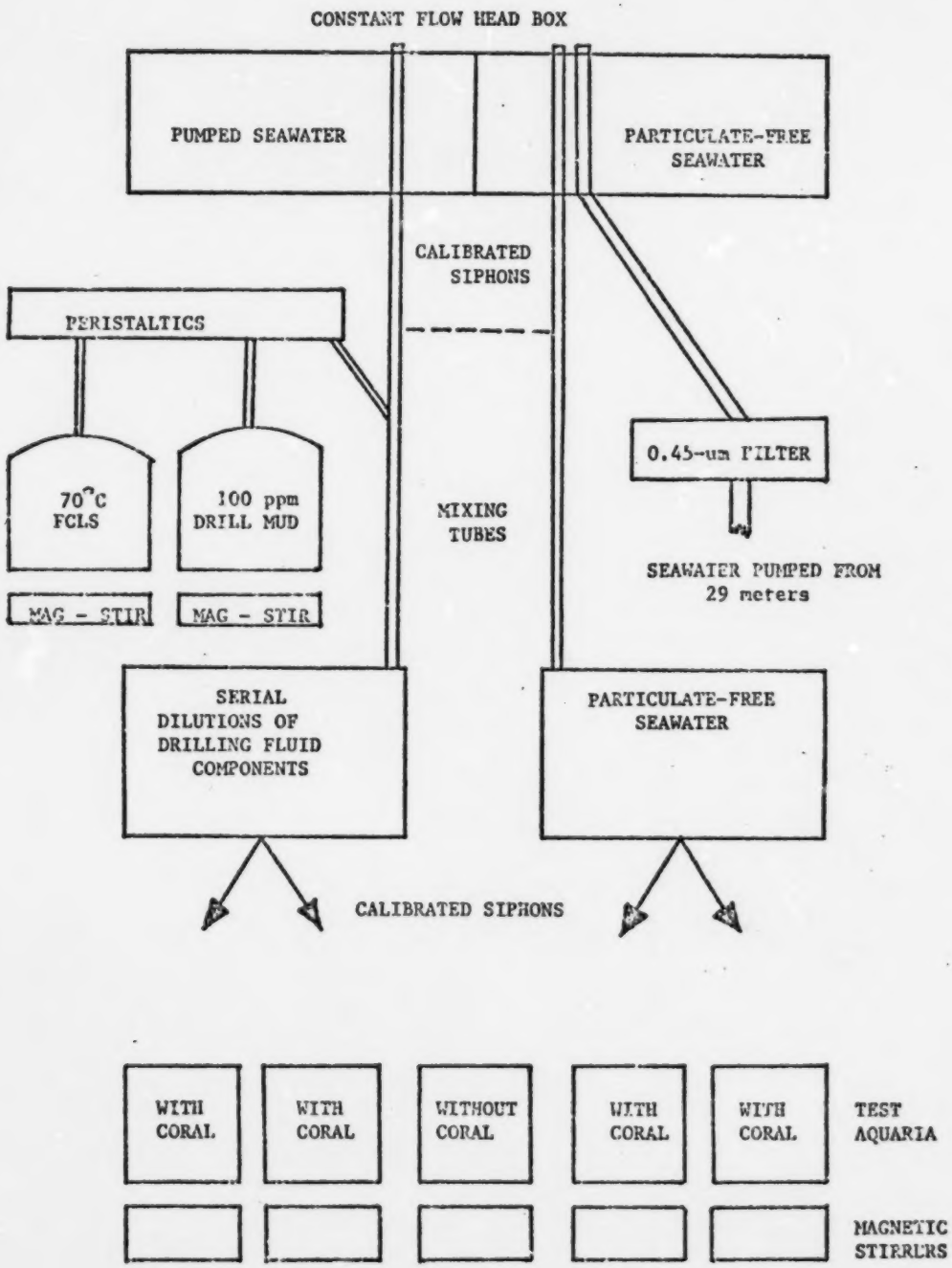
Figure 2. Locations of known coral reefs and zones of limited coral growth within the Gulf of Mexico. After Smith, 1954; Jordan, 1962; Logan, 1969; and others.

mixed by a magnetic stirrer. I allowed the corals 48 hours to acclimate to the circulation regime of the smaller chambers and to recover from incident handling (Days 1 and 2). All corals appeared healthy (polyps expanded and actively feeding) within 45 minutes after placement in the experimental aquaria.

Experimental Design. Sixteen corals were arrayed randomly (using a random number table) among four series (Table A-2, Appendix). Each series consisted of five test chambers: four with corals and the one coral-free system control. Corals in series 1 were exposed to raw seawater uncontaminated with either drilling mud or FCLS. Corals in series 2 were exposed to a 100 ppm suspension of drilling mud collected 3 April 79 from a working drill rig in Mobile Bay, Alabama. Complete chemical analysis of this drill mud is not yet available, but it was selected for this series of experiments since it was supposed to have a low FCLS concentration. Corals in series 3 and 4 were exposed to this 100 ppm drill mud spiked with 3 ppm and 10 ppm FCLS, respectively. FCLS was obtained from the Baroid Petroleum Services/NL Industries, Inc., trade name--Q-Broxin.

The introduction of drill mud and FCLS was carried out using a Sigma peristaltic pump and a two-channel Milton Roy peristaltic pump respectively (Figure 3). The FCLS was delivered to the drill mud suspension at 70^{o} centigrade to approximate down-well conditions. This mixture had cooled to ambient temperature, however, before being introduced into test aquaria. All peristaltic pumps were calibrated twice daily during the course of the experiment. FCLS-drill mud stocks were changed daily to minimize bacterial growth and chemical aging.

Figure 3. Diagram of the system used in this study.



Twice daily (noon and 4:00 PM) the flow-through aquaria were shut down for two-hour monitoring periods. During this time, circulation within the aquaria was maintained by the magnetic stirrers. Ammonium excretion was measured by difference between initial and final concentrations using a modified Solorzano method (Biggs, 1977) run on site using a Bausch and Lomb digital spectrophotometer (model 710). For ammonium analyses, 75 ml of seawater were withdrawn from each aquarium and filtered through 0.45 um GFC filters. To minimize contamination each filter was prerinsed with 25 ml of sample and a new filter was used to sample each aquarium. The phenol-alcohol reagent was added immediately to the filtrate and the remaining reagents within two hours of sampling. Color development was carried out in the dark, to avoid photochemical oxidation (Deggobis, 1973). After 5-12 hours, absorbance was measured in 5 cm cuvettes at 640 nm. New standard curves were constructed daily by spiking filtered seawater with 0, 2, 4, and 6 ug-at NH_4Cl/liter.

Oxygen consumption was also calculated by difference between initial and final concentrations determined polarographically using an Orbisphere model 2604 multichannel oxygen instrument. The probes were individually calibrated against water-saturated air between each measurement period to compensate for any drift. At intervals throughout the experiment, time-course respiration of individual corals was monitored by connecting probes to a multichannel recorder. As expected, respiration rates were linear throughout the measurement interval and agreed with oxygen uptake rates calculated by difference.

Prior to each measurement period, suspended particulates were displaced from each test chamber with a 4-5 minute flush of 8-10 liters of

0.45 um filtered seawater. This minimized extraneous chemical or biological changes in oxygen and ammonium in the test aquaria and allowed coral metabolic responses to be isolated for study. Coral metabolic responses were further isolated by subtracting the appropriate treatment control aquarium from the individual coral metabolic responses. To confirm that this flush displaced the bulk of the 100 ppm drill mud suspension, particulates remaining in aquaria after the flush were collected on a 0.45 um Millipore filter and the flush effectiveness determined by difference. All surfaces of the experimental aquaria were scrubbed approximately every four days to control fouling.

Three banks of fluorescent lights were positioned approximately two feet above the series of test aquaria. Light levels were measured with a Nikon light meter and reduced using opaque diffusers to levels characteristic of the Flower Gardens at 30 meters. These fluorescent banks were automatically timed to turn on and off at 8:00 AM and 8:00 PM, respectively. A black Visqueen hood isolated this study from other experiments being run on the Stage-1 sea tables.

After 17 days of exposure to drill mud + FCLS regimes, drill mud + FCLS additions were discontinued and the surviving corals allowed 48 hours to recover in uncontaminated seawater (days R1 and R2). Corals were then weighed, placed in 100 ml of 1N NaOH and transported frozen to College Station, Texas for subsequent protein analysis (Lowry et al, 1951). All oxygen and ammonium data have been reported on a protein basis to standardize differences in coral size, morphology, and calcification.

Salinity, temperature, laboratory seawater pH, pH of the drill mud + FCLS suspensions, and general weather conditions were monitored daily. Every night corals were examined, using a dim red-filtered light, for

approximate percentage of polyp expansion, feeding activity, polyp coloration, and aberrant behavior (i.e. mesenterial filament extrusion).

RESULTS

Flush of Particulates. Trials in which two aquaria containing flow-through 100 ppm drill mud suspensions were flushed with 0.45 um filtered seawater prior to closing the systems showed that 8.2±0.3 mg of particulates remained after a one-minute flush (flush rate 120 liters $hour^{-1}$). Since the pre-flush concentration of suspended drill mud was 100 mg $liter^{-1}$, I calculate that such a flush removed 92% of the particulates. In practice, test aquaria were individually flushed for up to 4-5 minutes prior to each measurement interval. Hence, flushing reduced the concentration of suspended particulates by at least twelve-fold prior to determination of oxygen consumption and ammonium excretion.

Adequacy of Controls. During each two-hour measurement period, changes in ammonium and oxygen levels in pre-flushed control aquaria (those without corals) averaged only -0.06 ml O_2 $liter^{-1}$ (s.d. = ±0.04, n=80) and 0.01 ug-at NH_4^+ $liter^{-1}$ (s.d. = ±0.25, n=80). Table A-3 in the Appendix indicates the mean daily changes in oxygen and ammonium utilization by these control aquaria. These changes were consistently low and were uncorrelated with the previous drill mud-FCLS regimes in all series. A standard test for chemical oxygen demand, using 250 ml ground glass stoppered bottles, confirmed that there was no significant COD for any experimental combination of drill mud + FCLS within a three hour period. Data (following) are expressed as ml O_2 $liter^{-1}$ (± measured range, n=2):

Table 1. Results of the chemical oxygen demand test.

Treatment	Initial O_2	1 Hour Later	3 Hours Later
Filtered SW	5.6±0.1	5.6±0.1	5.6±0.1
100 ppm DM	5.6±0.1	5.6±0.1	5.5±0.1
DM + 3 ppm FCLS	5.6±0.1	5.6±0.1	5.4±0.1
DM + 10 ppm FCLS	5.6±0.1	5.6±0.1	5.5±0.1

Coral Metabolism. Arithmetic averages of the mean hourly rates of ammonium excretion and oxygen consumption of the four corals exposed to each experimental regime are illustrated in Figures 4 and 5. In general, during the first week of the experiment, corals responded to increased drill mud + FCLS stress by increasing their ammonium excretion and oxygen consumption. Corals reached their highest average rates of excretion and respiration by the end of the first week of continuous exposure. Rates then decreased during the next week, and, after a secondary increase in excretion and respiration between days 10-13 most pronounced in those corals exposed to FCLS spikes, leveled off to near-initial rates by the end of the second week.

Two corals, one exposed to 100 ppm drill mud + 3 ppm FCLS and the other to 100 ppm drill mud + 10 ppm FCLS, became noticeably moribund as the experiment entered its second week. These had exhibited the most rapid and most consistent increases in excretion and respiration during the first week of exposure to FCLS. By week two, their polyp expansion was dramatically reduced, and each was removed from the experiment when areas of bare corallum suggested the onset of polyp death. While all corals exposed to FCLS reacted by reducing their polyp expansion behavior, only the two cited above showed mass mortality of polyps.

Figure 4. Ammonium Excretion - Arithemetic averages of the mean excretion rates of four corals exposed to each treatment (ug-at NH_4^+ g^{-1} coral protein h^{-1}).

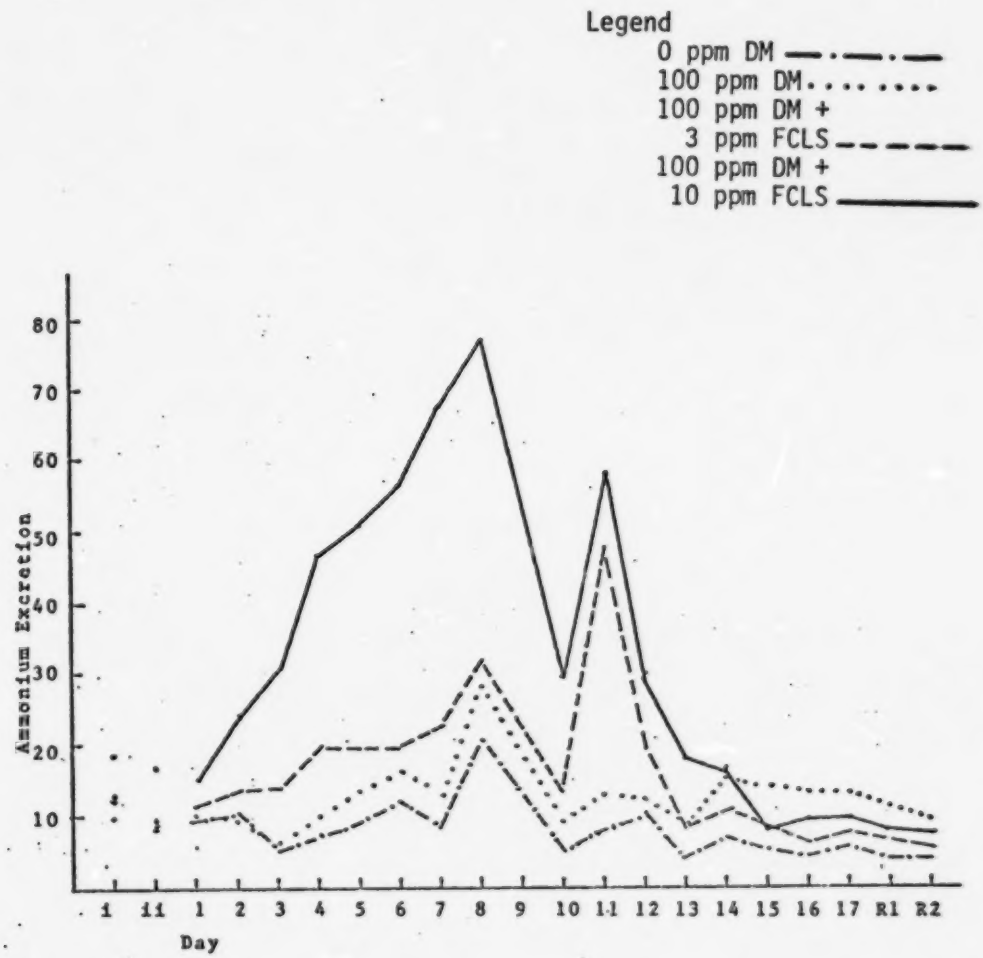
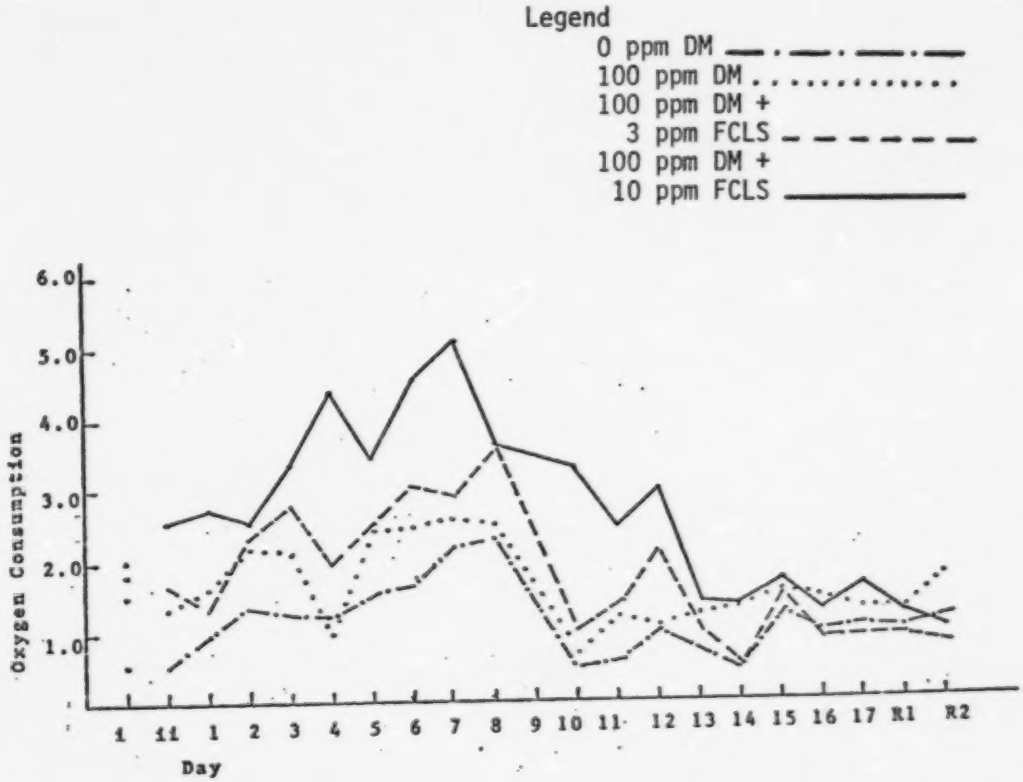


Figure 5. Oxygen Consumption - Arithmetic averages of the mean respiration rates of four corals exposed to each treatment. Expressed as ml O_2 g^{-1} coral protein h^{-1}.



Experimental and Individual Variability. Oxygen consumption and ammonium excretion of individual corals were less variable between measurements made twice daily compared with those made on successive days or between individuals (see Appendix, Figures A-1 and A-2, Tables A-4 and A-5). No consistent diel differences were noted between morning and afternoon measurements although between-individual variability was greatest at the highest FCLS exposure and least among corals exposed to drill mud only or to uncontaminated seawater. For instance, ammonium excretion of individual corals exposed to drill mud + 10 ppm FCLS varied by up to 230 ug-at NH_4^+ mg^{-1} protein $hour^{-1}$, or an order of magnitude greater than that of corals in uncontaminated seawater (Table A-5, day 8).

As emphasized in the preceding section, corals exhibited a net increase in oxygen consumption and ammonium excretion during the first week of the experiment, followed by a net decrease in these parameters during the next week. Although these trends were most pronounced in the respiration and excretion of corals exposed to the highest drill mud + FCLS levels, they were also present among corals in uncontaminated seawater. Moreover, the individual daily increases or decreases in respiration and excretion rates contributing to these trends were in phase among most of the experimental animals. For example, 14 out of the 16 corals showed a sharp decrease in oxygen consumption between days 3-5, which was contrary to the overall trend of net increase in respiration during the first week (Figure A-1). Furthermore, 13 of the 16 showed a temporary sharp increase in respiration and excretion between days 10-11 before these parameters declined to near-initial levels by day 14. These changes in respiration and excretion were not correlated with changes in temperature, salinity, or pH in the aquaria during the period of our

experiment, which remained effectively constant at $25^{o}\pm1^{o}C$, $35.0\pm0.5^{o}/oo$ and pH of 8.25 ± 0.05, respectively.

It is suggested that this phased variability in respiration and excretion possibly reflected stochastic changes in ration and/or digestive efficiency which were superimposed on the corals' metabolic responses to drill mud + FCLS. For example, food availability in experimental aquaria depended on its continued presence in the raw seawater pumped from a depth of 29 meters to a common head tank by the laboratory seawater system. It is conceivable that zooplankton and other potential food particles may have been present near the seawater intake at lower than average or higher than average densities between days 3-5 and days 10-11, respectively, and so correspond to the unexpected metabolic signatures.

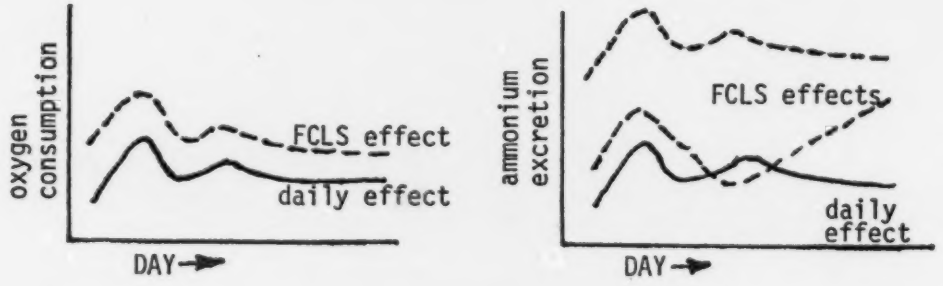
Analysis of variance: Table 1 presents results of a one and two-way analysis of variance of oxygen consumption and ammonium excretion of corals through day 17 of this experiment. Corals whose rates of respiration and excretion were aberrant from the others in each series were excluded from analysis. These included the two corals in FCLS enriched treatments which subsequently became moribund and were removed prior to the termination of the experiment and one coral uncontaminated by either drill mud or FCLS. The biomass of this latter coral was 2-3 times greater than any of the others (Table A-2), and consistently exhibited aberrantly high O-NH_4-N ratios (see discussion).

F values for remaining corals which were calculated for a one-way analysis of variance indicated that significant variability ($p<.0002$) occured among the individual corals in their rates of oxygen consumption and ammonium excretion within each series, when considered over the 17 day experimental period (Table 1, one-way analysis of variance). Analysis indicated that this between-coral variability was equally significant over shorter experimental periods (specifically days 1-7, days 8-13, and days 14-17). Figures A-1 and A-2 in the appendix document these individual differences in metabolism.

Variability in oxygen consumption and ammonium excretion between successive days (see DAY, Table 1) was highly significant ($p<.0001$). Variability between days accounted for 50% of the total variability within the sums of squares for oxygen consumption and 27% of the total variability for ammonium excretion. Note from figures A-1 and A-2 that in addition to daily changes in amplitude of metabolism, these changes were frequently in phase among corals in all series.

A treatment-day interaction was included to assess the interaction

betweem FCLS enrichment and the daily variability in oxygen consumption and ammonium excretion (see TRT*DAY, Table 1). The interaction of days and treatments was highly significant for excretion ($p<0.001$) although not significant for oxygen consumption ($p=0.23$). I interpret this to indicate that any differences in respiration resulting from FCLS enrichment were masked by the observed daily oscilations in oxygen consumption between treatments. Conversely, the daily oscilations in ammonium excretion between treatments were either of insufficient amplitude to mask changes resulting from FCLS enrichment or were out-of-phase with the latter. This hypothesis can be expressed by the following hypothetical graphs:



In summary, I suggest that it is this strong day effect coupled to the observed individual variability of corals within treatments which masked any statistically significant differences in oxygen consumption between corals exposed to different FCLS enrichments (see TRT, Table 1).

The effects of FCLS enrichment on the ammonium excretion of corals between treatments was isolated by a Duncan's multiple range test (Steele and Torrie, 1960). This procedure confirmed that differences in ammonium excretion significant at the $p=0.10$ level existed between corals exposed to uncontaminated seawater and the corals exposed to 100 ppm drill mud + 3 ppm FCLS.

Table 2: One and two-way analysis of variance results (days 1-17, excluding aberrant corals).

<u>One-way analysis of variance</u> to test variability of animals within treatments.

Series 1: uncontaminated control (n=3)
oxygen consumption - $F = 15.86$, $p < f$ 0.001
ammonium excretion - $F = 8.40$, $p < f$ 0.0008

Series 2: 100 ppm drill mud (n=4)
oxygen consumption - $F = 6.77$, $p < f$ 0.0006
ammonium excretion - $F = 8.26$, $p < f$ 0.0001

Series 3: 100 ppm drill mud + 3 ppm FCLS (n=3)
oxygen consumption - $F = 19.90$, $p < f$ 0.0001
ammonium excretion - $F = 5.52$, $p < f$ 0.0023

Series 4: 100 ppm drill mud + 10 ppm FCLS (n=3)
oxygen consumption - $F = 61.94$, $p < f$ 0.0001
ammonium excretion - $F = 43.00$, $p < f$ 0.0001

<u>Two-way analysis of variance</u>

Dependent Variable: Oxygen Consumption

SOURCE	DF	SUMS OF SQUARES	MEAN SQUARES	F	Pr>F
TRT	3	13.4582	4.48606	1.67	0.2420
AN(TRT)	9	24.1250	2.68056	10.46	0.0001*
DAY	15	62.0880	4.13920	15.99	0.0001*
TRT*DAY	45	13.6876	0.30417	1.19	0.2260
ERROR	135	34.6000	0.25629		

Dependent Variable: Ammonium Excretion

SOURCE	DF	SUMS OF SQUARES	MEAN SQUARES	F	Pr>F
TRT	3	2381.597355	793.85677	2.70	0.1084*
AN(TRT)	9	2638.296875	293.14409	7.13	0.0001*
DAY	15	6163.365384	410.89102	10.86	0.0001*
TRT*DAY	45	5286.806490	117.48459	2.86	0.0001*
ERROR	135	5551.453125	41.12188		

LEGEND: TRT=TREATMENT, AN(TRT)=ANIMALS WITHIN TREATMENTS, DAY=DAYS 1-17, and TRT*DAY=INTERACTION OF TREATMENT AND DAYS.

DISCUSSION

Sensitivity of Madracis to FCLS. Thompson and Bright (1977) reported that Madracis mirabilis corals exposed in the laboratory to combinations of drill mud and FCLS demonstrated alterations in normal polyp expansion behavior. Using time-lapse cameras to monitor the percentage of polyps expanded and searching food, they found that corals exposed to drill mud suspensions of 1 ppt or greater showed markedly reduced percentages of polyps expanded, relative to controls uncontaminated by drill mud. Corals were more sensitive to FCLS than to drill mud. Those exposed to dissolved FCLS levels of 100 ppm died within five days, which suggested that these animals were more sensitive to FCLS than the other marine organisms examined to date (see Table A-1, Appendix).

Thompson (1977) also reported that M. mirabilis survived 29 days of continuous exposure to 100 ppm drill mud and to 10 ppm dissolved FCLS, but while drill mud at these concentrations had no visible behavioral effects, all corals exposed to 10 ppm FCLS showed curtailed polyp expansion.

To determine whether coral behavioral changes might have measurable metabolic analogs, Westerhaus (1978) monitored oxygen consumption and ammonium excretion of M. mirabilis corals subjected to four series of dissolved FCLS (75, 50, 10, and 0 ppm) added to raw seawater in flow-through aquaria at Stage-1 (field season 1977). She reported that oxygen consumption for FCLS-stressed corals, relative to corals in uncontaminated seawater, increased daily throughout the first three days of exposure but decreased on the fourth day. All corals exposed to 50 and

75 ppm FCLS died by the fourth day of exposure, and one of the two exposed to 10 ppm FCLS died after the seventh day.

Westerhaus' data were characterized by extremely high variance, relative to calculated mean levels of oxygen consumption and ammonium excretion. Accordingly, when tested by analysis of variance (Steele and Torrie, 1960), no differences significant at the 0.05 level could be demonstrated among the four series, days of run, or the cross product of series and days. Since Westerhaus did not flush her aquaria prior to shutting them down for determination of oxygen consumption and ammonium excretion, it is likely that natural particulates in suspension (such as bacteria, phytoplankton, detritus, and microzooplankton) and their buildup on aquarium walls masked coral respiration and excretion. In fact, changes in oxygen and ammonium in "control" aquaria (without corals) were frequently of the same magnitude or higher than those in experimental aquaria. When these "control" activities were subtracted, net uptake of ammonium was indicated in some of Westerhaus' experimental series.

To reduce changes in oxygen and ammonium in control aquaria, aquaria were redesigned for the present study to allow particulates to be flushed efficiently and to permit aquarium surfaces to be scrubbed to remove drill mud buildup and/or algal growth. The number of experimental replicates in each series was also increased from two corals to four to allow a more effective comparison between the variation in individual coral response with differences in response between series.

Not unexpectedly, M. decactis showed sensitivity to drill mud + FCLS similar to that reported by Westerhaus for M. mirabilis, since the two are congeneric species. Day-to-day increases in oxygen consumption which Westerhaus reported for M. mirabilis exposed to the highest FCLS

concentrations, followed by a sharp decrease to near-initial values, were demonstrated by M. decactis. Since the former species was exposed to higher levels of FCLS than those corals employed in the present study, the increase-decrease trend was compressed into four days for M. mirabilis compared to 14 days for M. decactis.

The higher polyp mortality recorded by Westerhaus, relative to that found in this study, may indicate: 1) the greater toxicity of dissolved FCLS, relative to FCLS added in conjunction with drill mud, 2) stress of accumulation of drill mud on corals when not flushed twice daily, or 3) interspecific differences in tolerance between M. mirabilis and M. decactis.

The present study failed to confirm the apparent ammonium uptake reported by Westerhaus. As suggested earlier in this report, failure to flush the natural particulates from Westerhaus' aquaria undoubtedly contributed to her apparent ammonium fluxes. Alternatively, ammonium release by the corals may have potentiated growth of an algal fouling community which then scavenged ammonium produced by the corals. Since nitrogen is frequently the nutrient in shortest supply in the marine environment (Ryther and Dunston, 1971), this nutrient is generally the limiting factor to marine algal growth. McCarthy et al. (1977) have demonstrated that many marine algae take up ammonium as a preferred nitrogen source, even at relatively low concentrations of NH_4^+. In the present study, I noted that fouling proceeded most rapidly in aquaria subjected to the highest FCLS concentrations, where aquarium surfaces had to be scrubbed approximately every three days.

Acclimation of Madracis to FCLS. Corals in the present study responded to increased FCLS stress by increasing their oxygen consumption and ammonium excretion, compared to corals in uncontaminated seawater. After two weeks, respiration and excretion rates declined to near-initial levels. Although respiration and excretion rates in corals exposed to FCLS apparently returned to near-initial levels after day 14, in these corals polyp expansion was markedly aberrant. Photographic documentation (see pictures in Appendix) of FCLS stressed corals revealed that increasing FCLS enrichment caused: 1) reduction in number of polyps expanded (i.e., little or no active feeding during either day or night), 2) extrusion of zooxanthellae (resulting in a banded coloration of the coral colony), 3) bacterial infections with subsequent algal overgrowths, and 4) large-scale polyp mortality in two of the colonies. In contrast, corals in uncontaminated seawater or those exposed to drill mud not enriched with FCLS actively fed and appeared "healthy" throughout the course of the experiment. Healthy Madracis polyps expanded up to 1 cm and exhibited tentacular "concerts" individually during the day and colonially at night.

During the recovery period (days R1 and R2), coral behavior improved dramatically within one day after drill mud + FCLS stress was discontinued. Except where tissue regression had progressed to bare corallum, activity in surviving corals increased so that near-initial percentages of polyps were expanded and actively feeding on day R2. This suggests that corals which survived exposure to FCLS were not moribund or metabolically exhausted, but could recover rapidly when stress was discontinued.

Sensitivity of Related Species to FCLS. During the 1978 summer field season at Stage-1, comparative studies of drill mud + FCLS stress were conducted using another scleractinian coral, Oculina diffusa. Thompson had previously recorded a 96-hour LD_{50} (acute static bioassay) of approximately 12,000 ppm for FCLS. In contrast to Madracis, Oculina showed no reductions in polyp expansion when exposed to 10, 25, and 50 ppm dissolved FCLS (Thompson and Krone, unpublished).

The differences in sensitivity between Madracis and Oculina correlate with the temperature tolerance ranges which each species exhibits. Madracis has a rather restricted temperature range of 11-27°C (Vaughan and Wells, 1943), whereas Oculina is more eurythermal (Thompson, personal communication). Oculina colonizes habitats ranging from shallow, nearshore jetties to the legs of offshore drilling platforms (including Stage-1) to deep water. Madracis has a more restricted geographic range than Oculina and seems restricted by the photosynthetic needs of its zooxanthellae to shallow reefs (Oculina has proportionally fewer zooxanthellae).

In general, tolerance ranges are influenced by the previous environmental history, age, sex, and reproductive status of an organism. When an animal approaches an environmental tolerance limit, homeostatic mechanisms must expend energy to maintain equilibrium. In general, exposure to near-threshold levels of one environmental parameter may decrease an organism's tolerance to another variable. The wider tolerance range which Oculina exhibits might thus allow it to be less susceptible than Madracis to xenobiotic stress (specifically, to stress induced by FCLS).

Corals and Zooxanthellae. Zooxanthellae are photosynthetic unicellular algae of the order Dinoflagellida which live endosymbiotically in the tissues of about 150 genera of invertebrates comprising eight phyla (Droop, 1963). Zooxanthellae are found in all hermatypic (reef-building) corals and many ahermatypic corals. They maintain a symbiotic relationship intracellularly in the gastrodermal cells of the host (Smith et al., 1969).

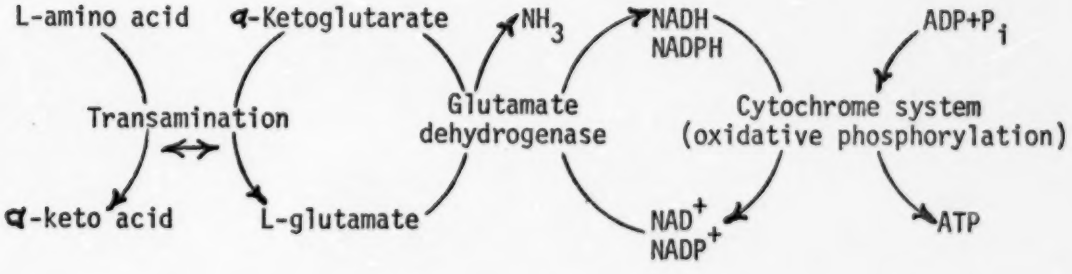
As endosymbionts, zooxanthellae have an immediate access to a source of carbon dioxide, they gain protection, and they may utilize organics and inorganics from their hosts for protein sources. McLaughlin and Zahl (1959) have found that zooxanthellae may utilize phosphate, phosphoric acid, nitrate, urea, uric acid, as well as guanine, adenine, and other amino acids generated by the host.

The hosts benefit as well from this symbiotic relationship. Corals may utilize photosyntate products produced by the algae and translocated to the coral host (Muscatine and Porter, 1977) and inorganic growth rates may be enhanced as well (Goreau and Goreau, 1959, 1961; Simkiss, 1964; Yamazato, 1966).

The symbiosis between Madracis decactis and its zooxanthellae determined the magnitude and direction of the net fluxes of oxygen and ammonium measured in my experiments. Inasmuch as I did not separate these endosymbiotic algae from their coral hosts, the ability of the zooxanthellae to take up NH_4^+ excreted by the corals coupled with possible differences in this uptake between stressed and unstressed zooxanthellae and probably extrusion of zooxanthellae by corals exhibiting banded coloration, all apparently contributed to variability in the metabolic parameters which were monitored in this study.

O/NH_4-N Ratios. A summary statistic which relates the number of oxygen atoms consumed to the number of nitrogen atoms excreted is a useful index of cellular metabolism (Harris, 1959). Because most aquatic invertebrates excrete ammonium as the principal end product of protein metabolism (Schmidt-Nielson, 1975), the whole-organism O/NH_4-N ratio is conveniently measured to approximate the general level of this cellular metabolism.

Since many marine invertebrates have low carbohydrate and lipid reserves, an increase in ammonium excretion should signal the transition from carbohydrate and lipid metabolism to catabolism of dietary or tissue protein. Accordingly, normal feeding-digestion cycles as well as stress may induce elevated ammonium excretion (Cates and McLaughlin, 1976). A biochemical pathway which incorporates ammonium excretion in invertebrates coupled to ATP production has been suggested by Pandian (1975).



The O/NH_4-N ratio, by atoms, for most marine invertebrates ranges from about 7 to 24 (Harris, 1959; Conover and Corner, 1968; Ikeda, 1974, 1977; Reeve et al., 1977; Biggs, 1977). For example, protein averages 16% nitrogen and requires 1.04 liters of oxygen for the complete combustion of 1 gram. If all the nitrogen is deaminated to ammonia, protein catabolism would be reflected by an O/NH_4-N ratio of about 6.8 (Ikeda,

1977). Oxidation of compounds low in nitrogen, like lipid or carbohydrate, would be reflected in a very high ratio (i.e. greater than 100). Oxidation of equivalent weights of protein and lipid requires 2.02 liters of oxygen for the complete combustion of 1 gram and yields an O/NH_4-N ratio of about 24 (Ikeda, 1974). Since few biochemical substrates have more than 16% nitrogen, O/NH_4-N ratios lower than 6.8 are generally interpreted as a breakdown of normal catabolism or excessive leakage of nitrogenous compounds (Biggs, 1977).

The coral with the consistently highest O/NH_4-N ratio also had the highest biomass (Table A-2 in the Appendix). This coral had approximately three times as much protein as other test corals in this experiment. Since experimental bias may have arisen due to the large size of this coral with respect to the one-liter experimental chamber, it was excluded from O/NH_4-N calculations and from analysis of variance of the oxygen consumption and ammonium excretion data (see page 20). Corals which subsequently became moribund were also excluded from O/NH_4-N computations and analysis of variance.

O/NH_4-N ratios for Madracis decactis in this study are generally in accord with those reported for marine invertebrates (Table A-8 and Figure A-3 in the Appendix) and suggest the predominance of protein catabolism. Ratios less than the theoretical minimum of 6.8 were posted by corals at intervals throughout the experiment (most notably during days 10-14) but were not restricted to FCLS enrichment. As a consequence of the phased changes in oxygen consumption and ammonium excretion (see page 18) O/NH_4-N ratios also fluctuated in phase.

Corals not exposed to drill mud or FCLS showed O/NH_4-N ratios ($\bar{x} \pm$ s.d.) of 13.3 ± 4.2 through the first week of the experiment, with those

exposed to drill mud or FCLS posting comparable ratios (17.5±6.6). O/NH_4-N ratios for all corals decreased between days 8-12, followed by a return to initial values after day 15. The low O/NH_4-N ratios between days 8-12 shown by corals in all series suggest that corals might have been leaking nitrogen. Such a situation might have resulted from the presence of lower than average food concentrations at this period in the Stage-1 flow-through seawater system.

Many coral species exhibit a recycling mechanism between the coral host and its endosymbiotic algae as a means of conserving nitrogenous nutrients (Lewis, 1973; Pomeroy et al., 1974; Muscatine and Porter, 1977). Recent work by Muscatine and D'Elia (1978) suggest that corals with symbiotic algae take up and retain ammonium when presented with normal diel light regimes. Their work suggests that uptake and retention of ammonium is enhanced by light and that exposure to several hours of daylight is sufficient to maintain net ammonium uptake by coral zooxanthellae throughout the night. Since uptake of ammonium by coral zooxanthellae would decrease net NH_4^+ excretion and increase coral O/NH_4-N ratios, activity of zooxanthellae might cause the magnitude of tissue protein catabolism to be underestimated. Figure A-5 indicates that the O/NH_4-N ratios of corals exposed to 10 ppm FCLS were in fact higher and more variable than those of other corals throughout most of the experiment.

If photosynthesis exceeds photorespiration, there should be a net increase in the O/NH_4-N ratios. Net photosynthetic activity by zooxanthellae will produce available oxygen, increasing the O/NH_4-N ratio:

$$CO_2 + 2\ HOH \rightarrow CH_2O + HOH + O_2 \qquad (1)$$

while net photorespiration will consume oxygen and release NH_4^+ (Keys et al., 1978), decreasing the O/NH_4-N ratio:

$$2\ \text{GLYCINE} + \text{HOH} \quad \text{SERINE} + CO_2 + NH_3 + 2\ H^+ + 2\ e^- \qquad (2)$$

Ecological Implications. Corals provide an extensive (15% of the shallow sea floor within the 0-30 meter depth range) and important tertiary substrate which houses and maintains complex, highly productive biocoenoces in areas often surrounded by oligotrophic water (Goreau, 1961; Johannes, 1976; Smith, 1978). Reef systems generate and trap particulate organic carbon, nitrogen-rich "flocs", and ammonium (Coles and Stratham, 1973; Muscatine and D'Elia, 1978; Smith and Marsh, 1973).

Experiments by Thompson and Bright (1977) demonstrated that corals were unable to remove loads of drilling mud when these exceeded about 1 ppt. If mud cannot be removed, the polyps eventually suffocate and die or are overgrown by opportunistic algae. The type of animals that settle and grow in a particular locality appears to be based upon the physical composition and organic content of the sediment (Thorson, 1957). Since bare coral rock is a preferred substrate for the settlement of coral planulae, if such areas become sediment covered or overgrown they are no longer suitable settlement areas for the planulae (Mileikovsky, 1970, 1971; Thorson, 1966). In addition, expansion of existing colonies may be hampered by surrounding sediment (Dodge, R. E., 1974). Changes in the composition and relative abundance of settling organisms in the coral reef system could alter interactions among resident animals and interrupt community stability and efficiency, characteristic of the coral reef system.

Corals must produce mucus in order to remove loads of drill mud; accordingly, elaboration of mucus is a energetic loss to the colony. Moreover, corals may experience "nematocyst fatigue" if individual nematocysts fire in response to particles in seawater, or during the regurgitation of mud ingested by the corals.

Although polyps may be expanded both day and night, our observations indicate that M. decactis increased its feeding activity at night (increased individual and colonial tentacular concerts). Recent field work suggests that such increases in feeding activity are in phase with nocturnal migrations of zooplankton which by day dwell in cavities within the reef (Porter and Porter, 1977). Accordingly, although Gulf of Mexico drilling platforms generally work around-the-clock when production is in progress, if drilling discharges were not pumped into the immediate vicinity of coral reefs at night, the corals might recover quickly enough from the stress of daytime sedimentation loads to benefit from the availability of greater-than-average food concentrations at night.

The two corals which exhibited the most rapid and most consistent increases in excretion and respiration during the first week of exposure to FCLS appeared to be infected with a white mucus sheath of bacteria. A hydrogen sulfide odor was also noted during the regular sampling of these subsequently moribund corals. The strict anaerobic bacteria of the genus Desulfovibrio reduces sulfate to hydrogen sulfide which is a potent toxin to most aerobic organisms (Jørgensen, 1977). Since such bacteria are common to the marine environment (Zobell and Rittenberg, 1948; Fenchel and Reidl, 1970; Jørgensen, 1977), the odor of hydrogen sulfide is speculated as evidence as to the character of the observed infection.

I suggest that the accumulation of drill mud on the bare coral rock

or interstitial spaces between colonies may have created a micro-environment conducive to Desulfovibrio growth. Bacterial growth could have been potentiated by the presence of mucus elaborated to remove drill mud.

Alternative Measures of Physiological Stress. Levels of internal amino acid pools and activities of enzymes which regulate nitrogen metabolism may serve as indices of physiological stress which could supplement the "whole animal" responses measured in the present study. Since most marine invertebrates have minimal carbohydrate and lipid reserves (Schmidt-Nielson, 1975), internal amino acid pools and/or body proteins are rapidly catabolized to combat stress. Ammonium excretion serves as an indirect indicator of this process, since NH_4^+ is the most common excretory product of such catabolism.

Glutamate dehydrogenase (GDH) is believed to be a key regulatory enzyme involved in intracellular deaminations (Pandian, 1975). Accordingly, increases in GDH activity should correlate with stress responses in invertebrates. If the composition and concentration of cellular free amino acid pools changes as a result of GDH activity, this may also serve as a useful index of stress. Both GDH and total amino acid analysis are independent of the availability of living animals and may be run on tissue collected in the field and kept frozen in liquid nitrogen until subsequent laboratory analysis.

Conclusions. The efficiency of FCLS as a thinning agent has been documented in the introduction to this report. However, I suggest that if economically feasible, it would be ecologically advantageous to substitute a thinning agent less toxic than FCLS into drilling muds. Alternatively, other less toxic metals might be substituted for the chromium

in FCLS. In the debate of over-regulation versus laissez-faire regulatory policies for offshore oil and gas resource production and development, such cost-benefit considerations may have far-reaching impacts.

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Table A-1. Results of toxicity studies of drilling mud and individual components on selected marine organisms.

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REFERENCE	MATERIAL TESTED	ORGANISM	RESULT (LC_{50}-96 hour)
NEKTON			
Monaghan (1977)	FCLS	Rainbow trout	2 ppt
Dames/Moore (1978)	FCLS	Onchrhychus gorbuscha (pink salmon fry)	3-29 ppt no effect after 4 days in cages near the discharge point
EG&G (1976)	drill mud	Menida menida (Atlantic silverside)	100 ppt
	FCLS		49 ppt
Dames/Moore (1978)	FCLS	Neomysis integer	74 ppt for a 48 hour LC_{50}
PHYTOPLANKTON			
EG&G (1976)	saltwater gel mud	Skeletonema costatum	100-1000 ppm
	FCLS		10-18 ppt
ZOOPLANKTON/ Crustacea			
EG&G (1976)	saltwater gel mud	Acartia tonsa	100-385 ppm
	FCLS		100-300 ppm
Dames/Moore (1978)	whole mud	Pandalus hypsinotus	32-150 ppt
Monaghan (1977)	7 drill muds	Anissogammurus confervicolus	10-50 ppt for a 48 hour LC_{50}
		Orchestia sp.	14-560 ppt
		Hemigrapsus nudus	53 ppt
METOFAUNA			
Alexander (1977)		Foraminifera	showed a significant decrease after drilling operations (t test=0.05), with the greatest decrease at 100 m and reduced populations to 1000 m
Pequegnat (1977)			showed an increase in the numbers of individuals between pre- and post-drilling samples, this increase was presumably due to natural seasonal variations which masked any potential effects caused by drilling
BENTHOS/ MACROFAUNA			
Knox (1978)	FCLS	Placopectin	1 ppt
Monaghan (1977)	drill mud	Modiolus modiolus	30 ppt with abnormal behavior at 3% but not 1% drill mud by volume
Dames/Moore (1978)		benthic infauna	no major effect on infauna at distances 100-200 m from the drill site

Table A-1. continued.

REFERENCE	MATERIAL TESTED	ORGANISM	RESULT (LC_{50}-96 hour)
BENTHOS/ MACROFAUNA			
Holland (1977)		benthic infauna	a decrease in numbers was reported between pre- and post-drill sites with no discernable change in populations at distances greater than 100 m
Targatz/Tobia (1978)		benthic infauna	colonization of estuarine benthic community was significantly less on sediment covered by barite or FCLS drill mud
Alexander (1977)	heavy metals	benthic infauna	no accumulations of heavy metals (Cd, Cr, Cu, Fe, Pb, Ni) were reported from macrofauna captured in trawls from the vicinity of drilling operations
Presely/Booth (1977)	heavy metals	benthic infauna	found basically the same conclusions as Alexander
HARD BANK COMMUNITIES			
Bright/Gerak (1978)		reef fish	epibenthic and demersal fish populations appeared healthy at post-drilling operations - no evidence of catastrophic mortalities
Thompson/Bright (1977)	used drill mud	Diploria strigosa Montastrea annularis M. cavernosa	used drill mud caused some mortality on three species of corals which were tested - the corals were unable to remove the used drill mud but all could remove the other three sediments which were tested
Westerhaus, (1978)	FCLS	Madracis mirabilis	observations indicated mortalities at 50 and 75 ppm FCLS - increased oxygen consumption and ammonium excretion appeared proportional to FCLS stress
Continental Shelf Associates (1978)		transects - no species available	no indications of positive or negative effects of the drilling activities on the health of monitored corals observed in situ
Marine Technical Services (1977)		transects - no species available	observations and photographic documentation revealed no discernable effects of the drilling operations on the health of the monitored corals in situ

APPENDICES

Photographs of experimental aquaria, healthy and stressed corals.

Table A-1: Literature survey of toxicity studies of drilling mud and individual componts on selected marine organisms.

Table A-2: Corals used in oxygen consumption/ammonium excretion experiments.

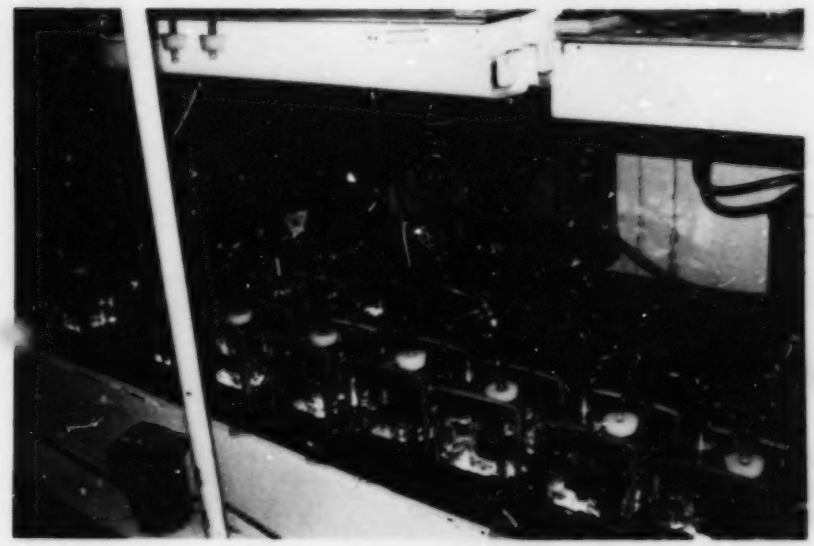
Table A-3: Average change in oxygen and ammonium concentrations in control aquaria after a one-hour measurement period.

Tables A-4 through A-7: Raw data - tables pertaining to the individual oxygen consumption and ammonium excretion of Madracis decactis, this study.

Table A-8: O/NH_4-N ratio for each drill mud + FCLS regime.

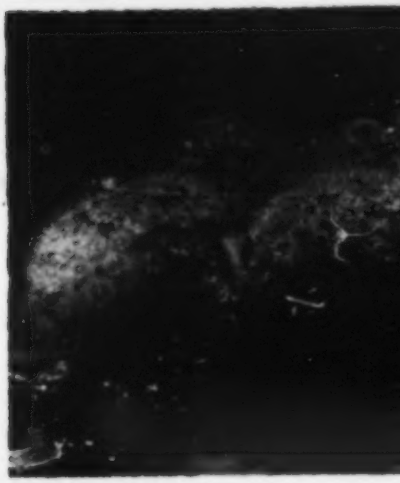
Figures A-1 through A-3: Raw data - figures pertaining to the individual oxygen consumption, ammonium excretion, and O/NH_4-N ratios of Madracis decactis, this study.

Experimental aquaria on the Stage-1 seawater tables-



Series 1: corals in uncontaminated seawate
expansion and the erect tentacle

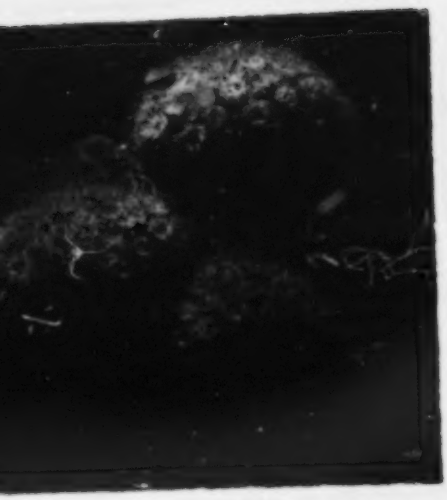
acclimation to smaller c



first day of experiment-

d seawater- (note the degree of polyp
tentacles)

smaller chambers-



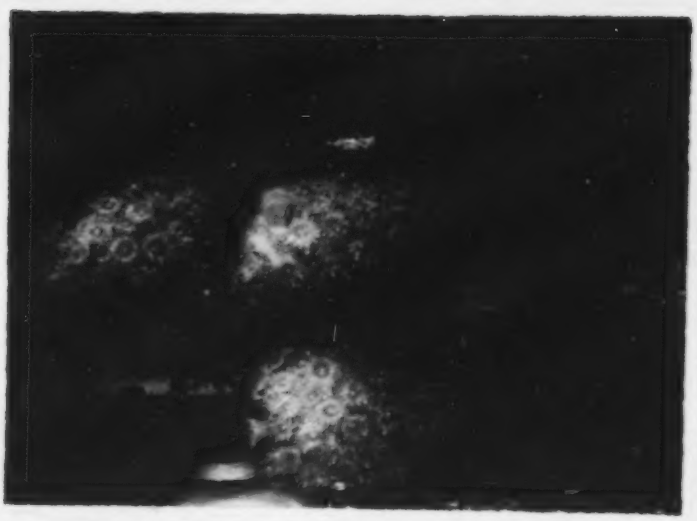
xperiment-

53<

Series 1 continued:

after two weeks-

recovery period-



Series 2: corals in 100 ppm drill mud- (note differences in polyp expansion, coloration, and apparent recovery)

acclimation to smaller chambers-

first day of experiment-

Series 2 continued:

after two weeks-

recovery period-

Series 3: corals exposed to 100 ppm drill mud + 3 ppm FCLS- (note the lack of tentacular activity, bacterial infection, and tissue regression)

acclimation to smaller chambers-

first day of experiment-

Series 3 continued:

after one week (note the bacterial infection)-

subsequent tissue regression where bacterial infection had occured-

Series 4: corals in 100 ppm drill mud + 10 ppm FCLS- (note the increasing degree of tissue regression)

acclimation to smaller chambers-

first day of experiment-

TABLE A-2. Corals used in oxygen consumption/ammonium excretion experiments.

Treatment	Protein (mg)	Skeletal Weight (g)
0 ppm Drill Mud	157	40.53
	76	25.66
	457 *	83.78
	194	31.38
100 ppm Drill Mud	170	25.03
	152	20.00
	135	34.34
	225	39.37
100 ppm Drill Mud + 3 ppm FCLS	126	37.34
	76	28.34
	187	77.07
	97	29.00
100 ppm Drill Mud + 10 ppm FCLS	140	44.26
	170	47.57
	164	55.66
	135	37.10

Table A-3. Average change ($\bar{x}$ ± s.d., n=4) in oxygen and ammonium concentrations in control aquaria (without corals) after a one-hour measurement period. Prior to each measurement period, aquaria were flushed with 0.45-um filtered seawater to remove the drill mud-ferrochrome lignosulfonate suspension so that it and natural particulates present in the aquaria during the normal flow-through mode did not mask coral metabolism. Positive values denote net production of oxygen and ammonium, negative values net consumption.

Day	Oxygen ml O_2 $liter^{-1}$ h^{-1}	Ammonium ug-at NH_4^+ $liter^{-1}$ h^{-1}
Initial day i	-0.04 ± .01	0.1 ± .2
Initial day ii	-0.03 ± .02	-0.3 ± .5
Day 1	-0.06 ± .01	-0.2 ± .5
Day 2	-0.08 ± .02	-0.1 ± .2
Day 3	-0.10 ± .01	0.1 ± .2
Day 4	-0.04 ± .02	0.1 ± .1
Day 5	-0.06 ± .01	-0.3 ± .6
Day 6	-0.10 ± .01	0.2 ± .4
Day 7	-0.13 ± .03	0 ± .2
Day 8	-0.17 ± .06	-0.2 ± .8
Day 10	0 ± .03	0.4 ± .4
Day 11	-0.03 ± .01	0.3 ± .4
Day 12	-0.08 ± .01	0.5 ± .2
Day 13	0 ± .01	0.4 ± .4
Day 14	0 ± .03	-0.1 ± .1
Day 15	-0.05 ± .03	0 ± .1
Day 16	-0.06 ± .01	-0.1 ± .1
Day 17	-0.03 ± .01	-0.3 ± .1
Recovery day 1	-0.06 ± .01	-0.2 ± .1
Recovery day 2	-0.06 ± .01	-0.1 ± .1
GRAND MEAN	-0.06 ± .04 n=80	-0.01 ± .25 n=80

Table A-4. Raw Data: Weight specific oxygen consumption of Madracis decactis (ml O_2 g^{-1} coral protein $hour^{-1}$). Values represent mean ± measured range of twice daily determinations except on days i, ii, 5, 13, 17, R-1, and R-2 when only single determinations were made.

Day	i	ii	1	2	3	4
0 ppm DM	12.2	1.0	1.0 ± .2	1.4 ± .1	1.3 ± 1.2	1.0 ± .6
	1.3	0.0	0.8 ± .1	2.9 ± .1	0.9 ± 1.2	1.0 ± .4
	1.5	1.1	1.2 ± .1	1.2 ± .1	1.6 ± .5	1.6 ± .2
	1.4	0.1	0.8 ± .1	0.7 ± .1	1.3 ± .1	1.2 ± .6
100 ppm DM	2.4	1.3	1.9 ± .2	2.4 ± .1	1.9 ± .1	1.7 ± 1.2
	1.3	0.5	1.0 ± .1	2.0 ± 1.0	1.9 ± .2	1.3 ± .4
	1.6	1.1	1.3 ± .1	1.7 ± .5	2.3 ± 2.3	0.5 ± .1
	2.8	2.6	2.4 ± .7	2.9 ± .1	2.8 ± 1.2	0.5 ± .2
100 ppm DM	0.8	1.3	1.0 ± .6	2.3 ± .2	2.1 ± .1	0.9 ± .2
+ 3 ppm	1.7	2.5	0.2 ± .4	2.5 ± .4	3.4 ± .1	2.3 ± .5
FCLS	2.1	2.4	2.7 ± .4	2.9 ± .2	3.5 ± .2	2.5 ± .4
	1.3	0.5	1.3 ± .3	1.(± .8	2.1 ± .3	2.2 ± 1.2
100 ppm DM	1.8	1.9	2.4 ± .8	2.0 ± .3	1.8 ± .1	2.3 ± .4
+ 10 ppm	0.1	2.3	5.1 ± .6	4.0 ± .1	6.7 ± .1	11.4 ± .6
FCLS	4.2	4.3	1.8 ± .1	2.4 ± .2	2.3 ± .2	1.8 ± .4
	1.1	1.7	1.6 ± .3	1.7 ± .3	2.5 ± .5	2.1 ± .6

5	6	7	8	10	11
1.3	1.6 ± .2	1.7 ± .1	1.6	0.4 ± .4	0.4 ± .2
1.9	2.2	3.5 ± .1	3.6	0 ± .4	0.4 ± .6
1.8	1.8 ± .1	2.4 ± .5	2.1 ± .6	1.4 ± .2	1.5 ± .2
1.1	0.9 ± .4	1.3 ± .5	1.8 ± .5	0.2 ± .5	0.3 ± .3
2.2	3.2 ± .2	2.8 ± .2	2.8 ± .5	0.8 ± .2	1.3 ± .2
1.4	1.8 ± .3	2.0 ± .8	1.9	0.6 ± .9	1.0 ± .3
2.1	2.1 ± .2	2.0 ± .6	1.7	0 ± 1.0	0.6 ± .2
3.8	2.9 ± .5	3.6 ± .1	3.8 ± 1.1	1.6 ± .1	2.0 ± .6
1.2	1.7 ± .2	2.3 ± .7	2.8 ± 1.0	0.4 ± .2	0.5 ± .1
2.7	3.1 ± 2.3	3.6 ± 1.7	3.4	0.4 ± .4	0.8 ± .3
4.1	4.6 ± .2	4.6 ± .3	5.3 ± .9	3.2 ± .2	3.7 ± .3
1.6	2.8 ± .1	1.0 ± .3	3.0	0 ± .3	0.6 ± .5
2.5	2.9 ± .4	2.9 ± .7	3.0 ± .4	1.6 ± .9	1.4 ± .1
8.8	9.7	12.6 ± 4.6	6.8 ± 1.8	9.6 ± 2.8	6.2 ± .8
o.1	2.7 ± .3	2.6 ± .6	2.1 ± .7	0.9 ± .9	1.0 ± .3
2.5	3.1 ± .2	2.3 ± .7	2.6 ± .3	1.0 ± .2	1.3 ± .2

Table A-4 continued.

12	13	14	15	16	17	R-1	R-2
0.4	0.5	0.4 ± .2	1.0 ± .2	0.6 ± .1	0.6	0.9	0.9
1.4 ± .4	0.6	0.4 ± .4	1.7 ± .8	1.4 ± .1	1.6	1.3	1.6
1.7	1.7	0.9 ± .2	1.5 ± .2	1.6 ± .1	1.7	1.6	1.6
0.6	0.3	0.3 ± .1	0.8 ± .1	0.6 ± .1	0.5	0.7	0.7
1.0 ± .6	1.6	2.0 ± .1	2.8 ± .2	2.7 ± .2	2.6	2.5	2.3
0.9 ± .2	0.9	0.9 ± .1	1.9 ± .1	1.2 ± .1	0.9	0.9	1.4
0.6 ± .2	0.8	0.8 ± .1	1.3 ± .5	0.9 ± .2	0.7	0.9	0.7
2.0 ± .1	1.8	1.7 ± .3	0.5 ± .1	1.0 ± .2	1.2	1.0	2.7
0.8 ± .1	0.6	0 ± .4	1.3 ± .8	0.6 ± .1	0.7	0.8	0.6
2.3 ± .4	1.7	1.0 ± .3	1.6	1.4 ± .1	1.4	1.3	1.1
4.4 ± .4	moribund	----	----	----	---	---	---
1.1 ± .1	1.0	0.5 ± .1	2.0 ± .3	1.0 ± .1	0.8	1.0	1.0
2.0 ± .2	1.3	0.6 ± .4	1.8 ± .9	1.0 ± .1	1.3	1.2	1.0
6.4 ± .3	2.3	3.1 ± 1.8	moribund	----	---	---	---
1.7 ± .1	0.9	0.8 ± .3	3.1 ± 1.3	1.7 ± .1	2.0	1.5	1.0
2.0 ± .1	1.3	0.7 ± .3	0.2 ± 1.0	1.3 ± .1	1.7	1.3	1.2

Table A-5. Raw Data: Weight-specific ammonium excretion of Madracis decactis (ug-at NH_4^+ g^{-1} coral protein $hour^{-1}$). Values represent mean ± measured range of twice-daily determinations, except on days i, ii, 5, 13, 17, R-1 and R-2 when only single determinations were made.

Day	i	ii	1	2	3	4	5
0 ppm DM	58	10	11 ± 4	9 ± 1	5 ± 1	8 ± 4	9
	8	47	14 ± 2	15 ± 5	7 ± 2	12 ± 6	14
	3	3	3 ± 1	4 ± 1	2 ± 1	3 ± 1	3
	4	6	8 ± 1	10 ± 1	5 ± 3	5 ± 1	8
100 ppm DM	3	10	9 ± 1	9 ± 1	6 ± 2	13 ± 1	14
	9	5	7 ± 1	5 ± 1	19 ± 4	6 ± 2	9
	10	11	12 ± 1	12 ± 3	18 ± 4	9 ± 3	9
	17	9	12 ± 1	14 ± 1	9 ± 1	13 ± 2	20
100 ppm DM	6	5	11 ± 2	6 ± 2	6 ± 5	6 ± 3	9
+ 3 ppm	11	10	18 ± 1	20 ± 1	19 ± 4	28 ± 17	20
FCLS	10	9	10 ± 1	12 ± 2	18 ± 4	25 ± 1	40
	21	10	13 ± 3	17 ± 11	9 ± 1	21 ± 8	9
100 ppm DM	6	3	10 ± 1	16 ± 3	6 ± 1	12 ± 2	10
+ 10 ppm	37	24	28 ± 1	62 ± 13	103 ± 8	162 ± 4	165
FCLS	2	5	13 ± 2	9 ± 1	5 ± 3	6 ± 2	4
	5	4	8 ± 1	8 ± 1	6 ± 4	8 ± 1	25

6	7	8	10	11	12
12 ± 1	8 ± 4	14 ± 3	5 ± 2	8 ± 2	12 ± 5
24 ± 9	13 ± 2	35 ± 3	6 ± 2	16 ± 8	17 ± 3
3 ± 1	3 ± 1	6 ± 2	3 ± 1	3 ± 1	3 ± 1
8 ± 1	10 ± 5	28 ± 15	4 ± 1	6 ± 1	8 ± 2
26 ± 2	14 ± 5	23 ± 6	5 ± 1	12 ± 3	13 ± 1
6 ± 2	6 ± 1	23 ± 15	6 ± 3	8 ± 3	6 ± 2
16 ± 9	10 ± 1	27 ± 13	9 ± 2	10 ± 1	11 ± 2
17 ± 2	21 ± 2	39 ± 15	16 ± 2	22 ± 2	18 ± 2
8 ± 1	8 ± 1	19 ± 7	6 ± 1	55 ± 2	13 ± 7
14 ± 1	18 ± 3	43 ± 17	16 ± 5	91 ± 2	20 ± 1
40 ± 3	53 ± 3	42 ± 25	24 ± 3	34 ± 1	31 ± 9
17 ± 4	12 ± 4	24 ± 4	6 ± 1	13 ± 1	17 ± 4
15 ± 2	20 ± 1	27 ± 9	11 ± 1	48 ± 1	8 ± 5
193 ± 39	232 ± 36	245 ± 17	95 ± 2	91 ± 2	86 ± 6
8 ± 1	10 ± 1	16 ± 8	5 ± 2	41 ± 1	12 ± 12
10 ± 5	8 ± 2	17 ± 3	7 ± 1	50 ± 1	10 ± 9

Table A-5 continued.

13	14	15	16	17	R-1	R-2
2	6 ± 2	6 ± 2	5 ± 1	5	3	3
6	13 ± 6	13 ± 6	8 ± 1	12	6	6
2	2 ± 1	2 ± 1	2 ± 1	2	2	2
5	6 ± 2	5 ± 2	3 ± 1	3	3	2
23	23 ± 1	28 ± 2	30 ± 1	29	21	14
3	9 ± 3	6 ± 3	5 ± 1	6	6	3
10	10 ± 2	5 ± 1	3 ± 1	4	4	6
18	18 ± 6	18 ± 7	15 ± 1	15	17	14
8	8 ± 2	7 ± 2	4 ± 1	5	4	3
10	14 ± 2	13 ± 13	8 ± 1	9	8	7
moribund	---	---	---	---	---	---
6	10 ± 1	5 ± 3	6 ± 1	9	6	5
4	10 ± 1	5 ± 1	6 ± 1	5	6	5
43	31 ± 2	moribund	---	---	---	---
20	13 ± 1	13 ± 6	16 ± 1	12	9	8
5	9 ± 1	7 ± 2	7 ± 2	12	10	10

Table A-6. Summary of weight-specific oxygen consumption (ml O_2 g^{-1} coral protein hour $^{-1}$). Values represent the arithmetic averages of the mean respiration rates of four corals exposed to each experimental treatment (± standard deviation, n=4).

Day	1	11	1	2	3	4
100 ppm DM	4.0 ± 5.5	0.5 ± 0.6	1.0 ± 0.2	1.6 ± 1.0	1.3 ± 0.3	1.2 ± 0.3
100 ppm DM	2.0 ± 0.7	1.4 ± 0.9	1.7 ± 0.6	2.3 ± 0.5	2.2 ± 0.4	1.0 ± 0.6
100 ppm DM + 3 ppm FCLS	1.5 ± 0.6	1.7 ± 0.9	1.3 ± 1.0	2.3 ± 0.5	2.8 ± 0.8	2.0 ± 0.7
100 ppm DM + 10 ppm FCLS	1.8 ± 1.7	2.5 ± 1.2	2.7 ± 1.6	2.5 ± 1.0	3.4 ± 2.3	4.4 ± 4.7
	5	6	7	8	10	11	12
	1.6 ± 0.4	1.7 ± 0.5	2.3 ± 1.0	2.3 ± 0.9	0.5 ± 0.6	0.6 ± 0.6	1.0 ± 0.6
	2.4 ± 1.0	2.5 ± 0.6	2.6 ± 0.8	2.5 ± 0.9	0.8 ± 0.7	1.2 ± 0.6	1.1 ± 0.6
	2.4 ± 1.3	3.1 ± 1.2	2.9 ± 1.6	3.6 ± 1.2	1.0 ± 1.5	1.4 ± 1.6	2.1 ± 1.6
	3.5 ± 3.8	4.6 ± 3.4	5.1 ± 5.0	3.6 ± 2.1	3.3 ± 4.2	2.5 ± 2.5	3.0 ± 2.3
	13	14	15	16	17	R-1	R-2
	0.8 ± 0.6	0.5 ± 0.3	1.3 ± 0.4	1.0 ± 0.5	1.1 ± 0.6	1.1 ± 0.4	1.2 ± 0.5
	1.3 ± 0.5	1.3 ± 0.6	1.6 ± 1.0	1.5 ± 0.8	1.3 ± 0.9	1.3 ± 0.8	1.8 ± 0.9
	1.1 ± 0.5	0.5 ± 0.5	1.6 ± 0.3	1.0 ± 0.4	1.0 ± 0.4	1.0 ± 0.3	0.9 ± 0.3
	1.4 ± 0.6	1.3 ± 1.2	1.7 ± 1.4	1.3 ± 0.4	1.7 ± 0.3	1.3 ± 0.2	1.0 ± 0.1

*One of the corals grew increasingly moribund and was removed between days 12 and 13; n=3.
**One of the corals grew increasingly moribund and was removed between days 14 and 15; n=3.

Table A-7: Summary of weight-specific ammonium excretion (ug-at NH_4^+ g^{-1} coral protein $hour^{-1}$). Values represent arithmetic averages of the mean excretion rates of four corals exposed to each experimental treatment (± standard deviation, n=4).

Day	0	11	1	2	3	4	5
0 ppm DM	18 ± 27	17 ± 20	9 ± 5	10 ± 5	3 ± 2	7 ± 3	9 ± 5
100 ppm DM	10 ± 6	9 ± 2	10 ± 2	10 ± 3	6 ± 3	10 ± 3	13 ± 5
100 ppm DM + 3 ppm FCLS	12 ± 6	9 ± 3	13 ± 3	14 ± 6	13 ± 6	20 ± 10	20 ± 14
100 ppm DM + 10 ppm FCLS	13 ± 16	9 ± 10	15 ± 9	24 ± 25	31 ± 49	47 ± 77	51 ± 76
	6	7	8	10	11	12	13	14
		9 ± 4	21 ± 13	5 ± 2	8 ± 5	10 ± 6	4 ± 2	7 ± 5
		13 ± 6	28 ± 8	9 ± 5	13 ± 6	12 ± 5	14 ± 9	15 ± 7
		23 ± 21	32 ± 12	13 ± 9	49 ± 33	20 ± 8	*8 ± 2	*11 ± 3
		68 ± 110	76 ± 113	29 ± 44	58 ± 23	29 ± 38	17 ± 18	16 ± 10
		57 ± 91	15	16	17	R-1	R-2	
		5 ± 2	4 ± 2	6 ± 4	4 ± 2	3 ± 2		
		14 ± 11	13 ± 12	13 ± 12	12 ± 8	10 ± 5		
		*8 ± 5	*6 ± 2	*8 ± 2	*6 ± 2	*5 ± 2		
		**8 ± 4	**8 ± 5	**10 ± 4	**8 ± 2	**8 ± 2		

* One of the corals grew increasingly moribund and was removed between days 12 and 13
** One of the corals grew increasingly moribund and was removed between days 14 and 15

Table A-8: O/NH_4-N ratio for each drill mud + FCLS regime ($\bar{x}$ + s.d.). Values represent the ratio of oxygen atoms consumed to NH_4-N atoms excreted. Three corals which consistently showed higher than average oxygen consumption and ammonium excretion were deleted from analysis.

Day	0 ppm DM	100 ppm DM	3 ppm FCLS 100 ppm DM	10 ppm FCLS 100 ppm DM
Day 1	7.4±2.0	14.7±4.3	6.0±4.3	17.2±4.5
Day 2	12.5±5.7	23.4±9.5	17.9±14.2	18.0±6.4
Day 3	19.3±6.8	19.1±10.3	22.7±7.8	36.2±8.9
Day 4	13.3±7.2	9.9±7.3	10.0±3.1	22.4±4.9
Day 5	12.4±0.4	11.2±7.8	13.3±2.3	11.1±10.2
Day 6	10.0±1.9	16.2±7.3	16.0±4.4	25.0±6.8
Day 7	18.2±6.2	20.2±6.5	17.0±9.2	20.6±6.8
Day 8	16.9±16.0	8.2±2.2	10.5±3.1	11.7±1.9
Day 10	3.9±3.6	8.0±5.9	2.7±3.0	13.9±1.9
Day 11	3.7±1.3	8.6±2.5	1.9±1.9	2.4±2.1
Day 12	5.7±2.3	8.8±3.7	7.2±2.7	17.6±4.9
Day 13	12.2±8.9	12.1±9.8	12.3±4.8	18.7±13.0
Day 14	4.4±1.6	10.1±6.8	3.6±3.3	5.9±0.8
Day 15	13.6±1.7	15.7±12.0	21.1±13.0	14.3±15.0
Day 16	14.7±3.6	15.5±10.0	14.6±1.1	13.7±3.7
Day 17	12.5±2.2	11.0±4.1	11.4±3.1	16.9±5.6
Recovery Day 1	22.3±4.0	12.3±6.2	15.7±1.8	14.8±3.1
Recovery Day 2	27.3±3.7	21.0±14.0	16.5±2.2	13.2±4.0
Treatment Grand Mean	11.3±5.0	13.3±4.7	11.8±6.4	16.6±7.8

Figure A-1. Oxygen Consumption of 4 experimental corals expressed as ml O_2 g^{-1} coral protein h^{-1}.

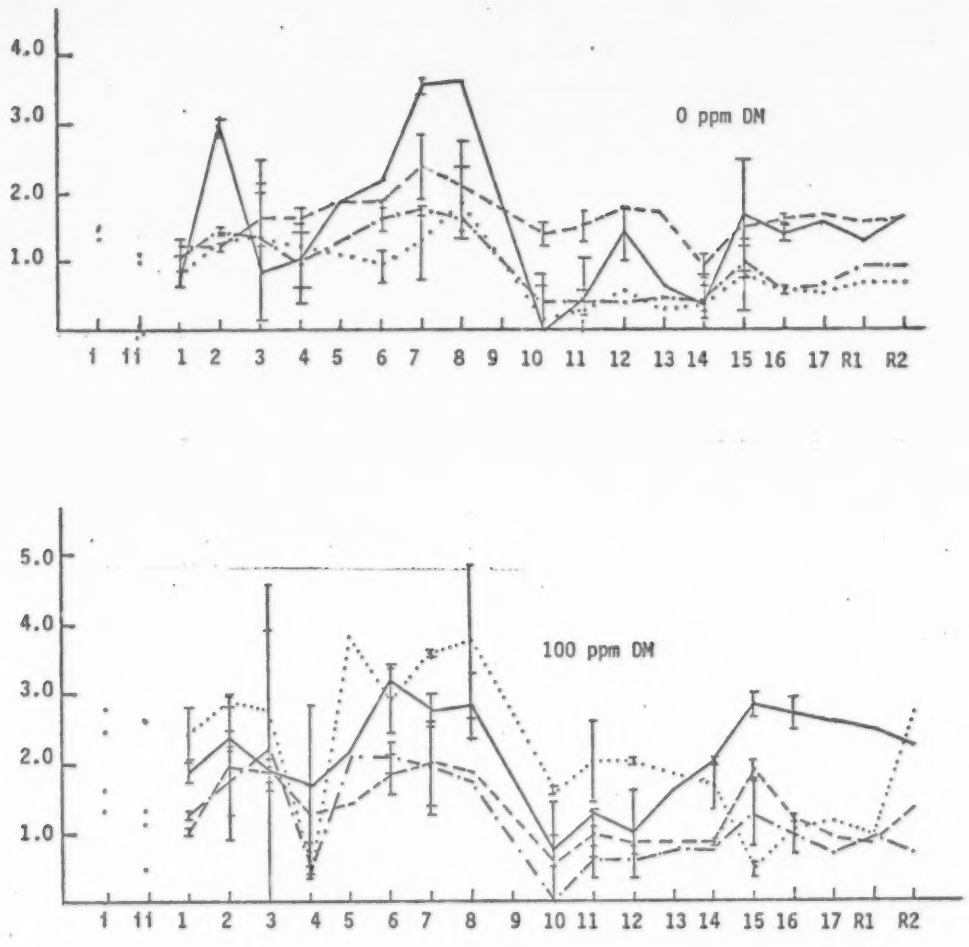
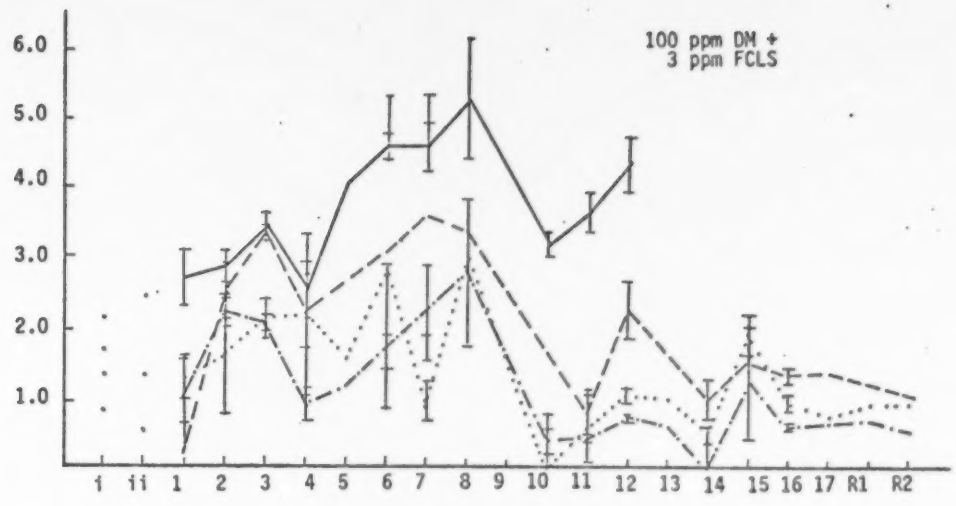


Figure A-1 continued.



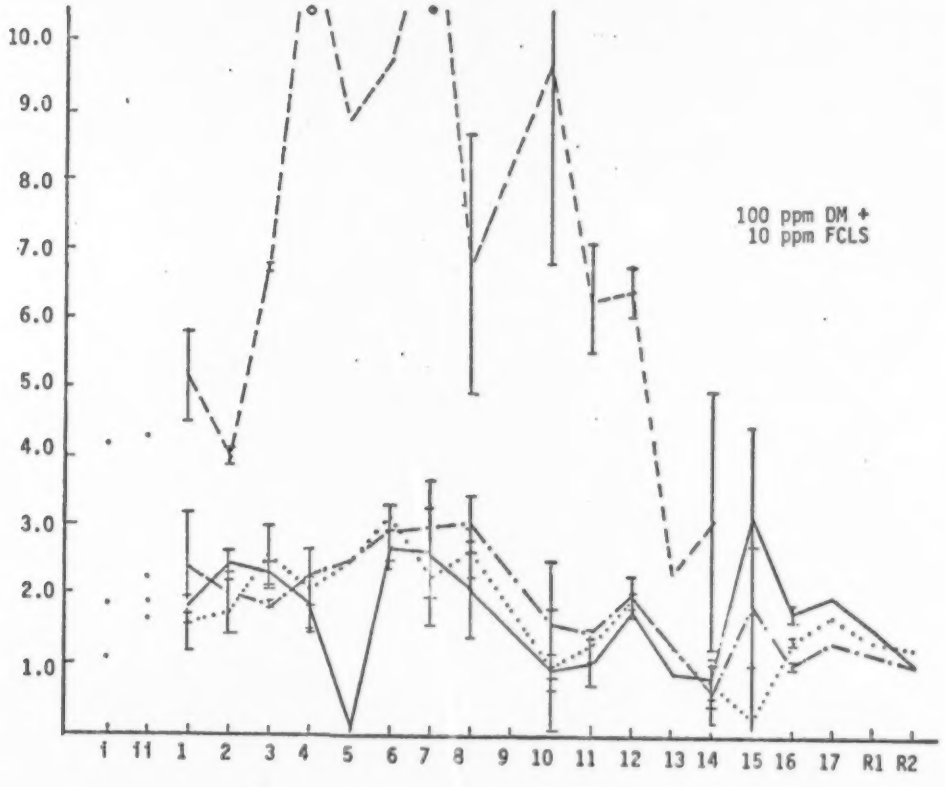


Figure A-2. Ammonium Excretion of 4 experimental corals. (ug-at NH_4^+ g^{-1} coral protein h^{-1})

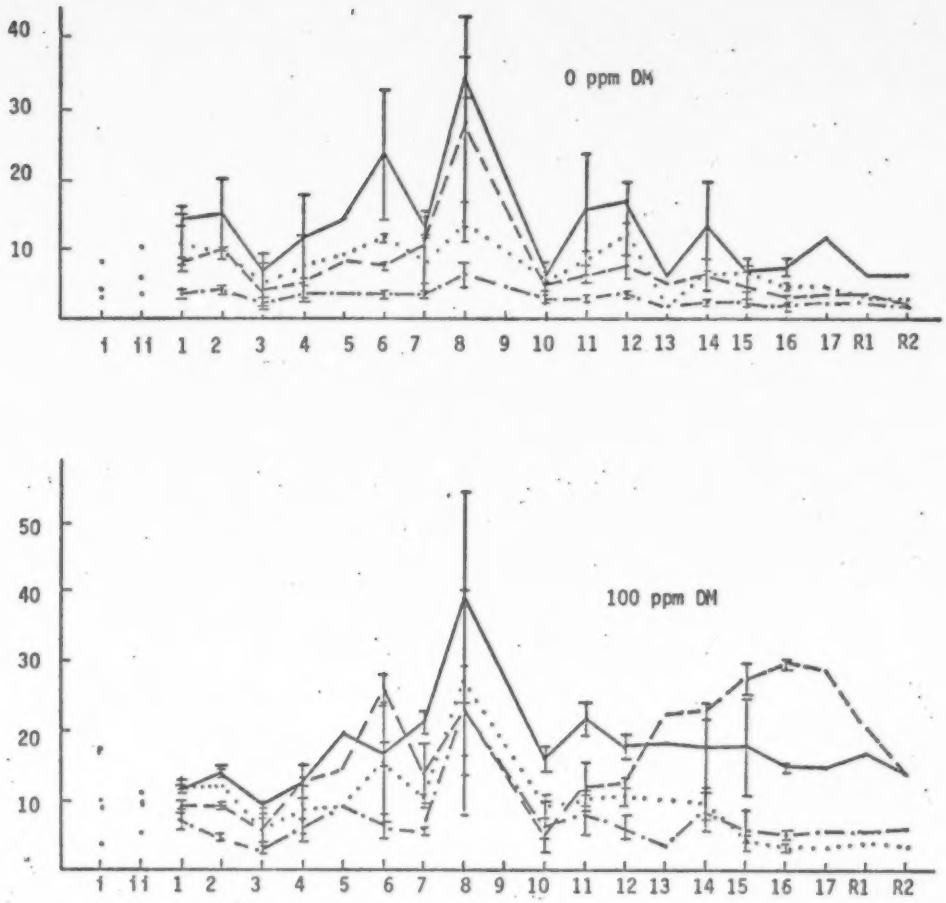
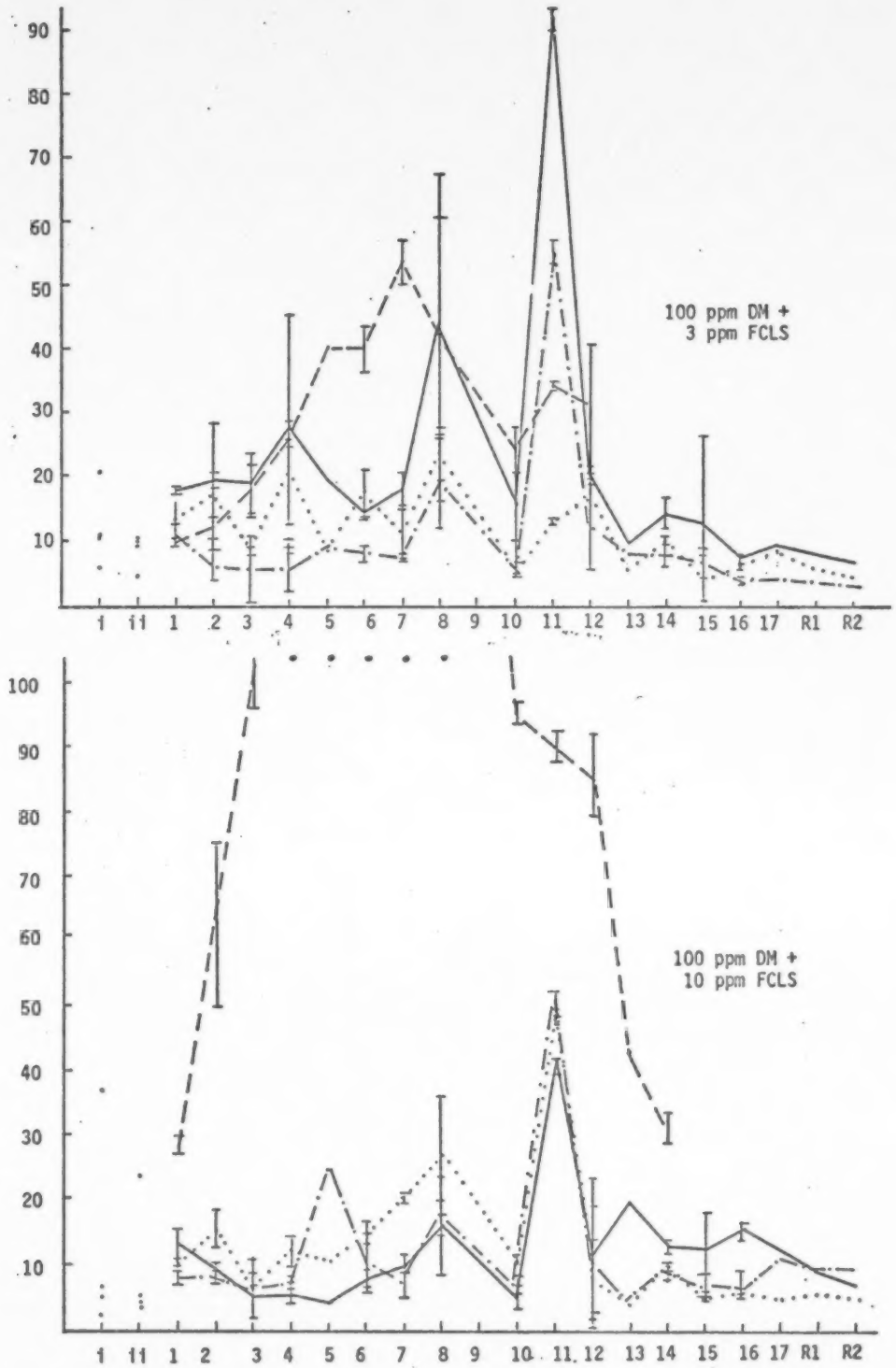


Figure A-2 continued.



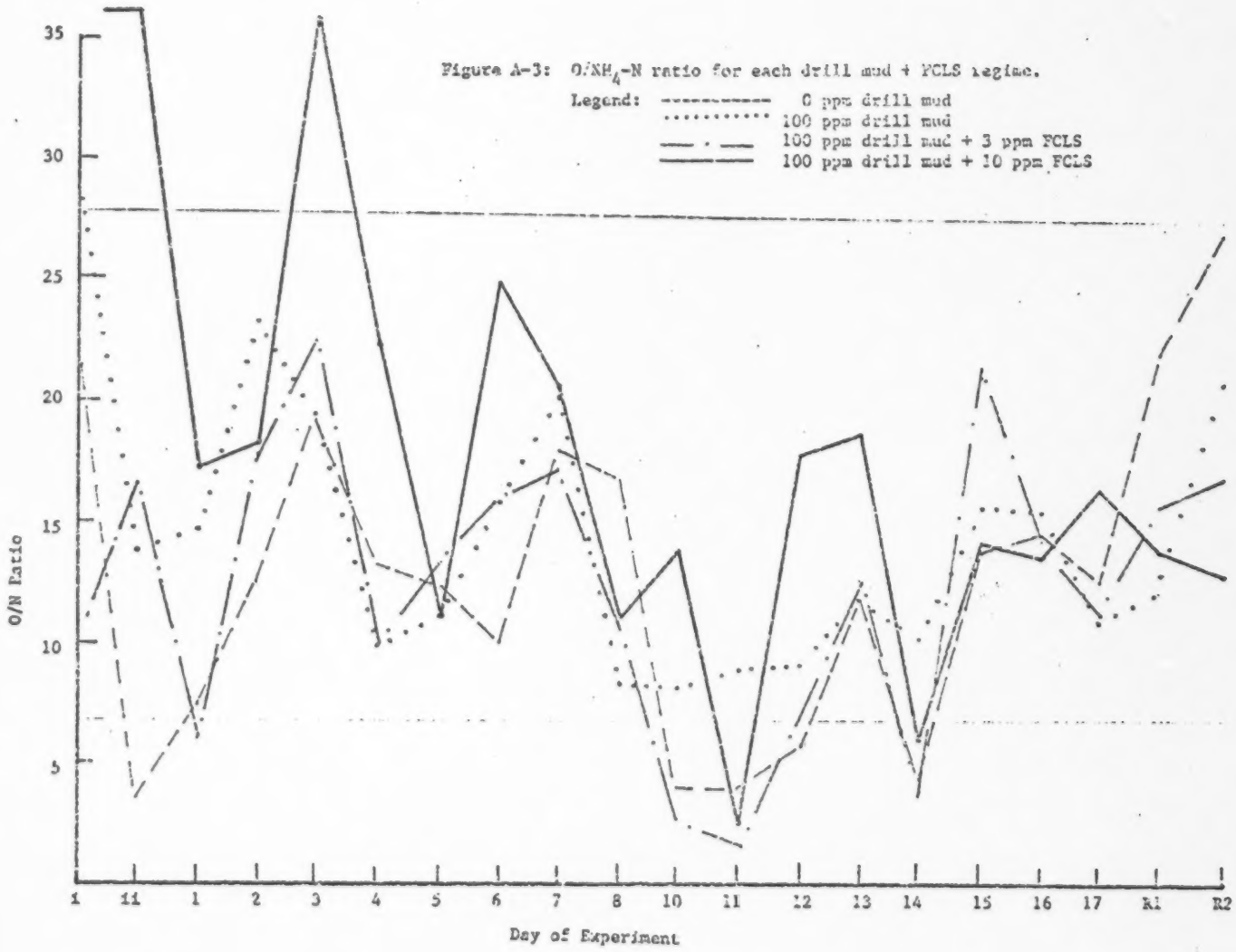


Figure A-3: O/NH_4-N ratio for each drill mud + FCLS regime.

Legend:
- ---------- 0 ppm drill mud
- ..... 100 ppm drill mud
- — · — 100 ppm drill mud + 3 ppm FCLS
- ——— 100 ppm drill mud + 10 ppm FCLS